METHODS OF USING AND COMPOSITIONS COMPRISING SELECTIVE CYTOKINE INHIBITORY DRUGS FOR THE TREATMENT AND MANAGEMENT OF MYELOPROLIFERATIVE DISEASES

1. FIELD OF THE INVENTION

This invention relates to methods of treating, preventing and/or managing myeloproliferative diseases and related syndromes which comprise the administration of selective cytokine inhibitory drugs alone or in combination with other therapies.

2. BACKGROUND OF THE INVENTION

2.1 PATHOBIOLOGY OF MPD

Myeloproliferative disease (MPD) refers to a group of disorders characterized by clonal abnormalities of the hematopoietic stem cell. *See e.g.*, *Current Medical Diagnosis & Treatment*, pp. 499 (37th ed., Tierney *et al.* ed, Appleton & Lange, 1998). Since the stem cell gives rise to myeloid, erythroid, and platelet cells, qualitative and quantitative changes can be seen in all these cell lines. *Id.*

MPD is further subdivided on the basis of the predominantly proliferating myeloid cell type. Erythrocyte excess is classified as "polycythemia rubra vera (PRV)" or "polycythemia vera," platelet excess as "primary (or essential) thromobocythemia (PT)," and granulocyte excess as "chronic myelogenous leukemia (CML)." A fourth subcategory of MPD is "agnogenic myeloid metaplasia (AMM)," which is characterized by bone marrow fibrosis and extramedullary hematopoiesis. *Cecil Textbook of Medicine*, pp. 922 (20th ed., Bennett and Plum ed., W.B. Saunders Company, 1996). These disorders are grouped together because the disease may evolve from one form into another and because hybrid disorders are commonly seen. Tierney *et al*, *supra*, at pp. 499. All of the myeloproliferative disorders may progress to acute leukemia naturally or as a consequence of mutagenic treatment. *Id.*

Most patients with PRV present symptoms related to expanded blood volume and increased blood viscosity. *Id.* at pp. 500. Common complaints include headache, dizziness, tinnitus, blurred vision, and fatigue. *Id.* The spleen is palpably enlarged in 75% of cases, but splenomegaly is nearly always present when imaged. *Id.* Thrombosis is the most common complication of PRV and the major cause of morbidity and death in this disorder. Thrombosis appears to be related to increased blood viscosity and abnormal platelet function. *Id.* Sixty percent of patients with PRV are male, and the median age at presentation is 60. It rarely occurs in adults under age 40. *Id.*

Thrombosis is also a common complication in patients suffering from PT. *Cecil Textbook of Medicine*, pp. 922 (20^{th} ed., Bennett and Plum ed., W.B. Saunders Company, 1996). A platelet count $\geq 6 \times 10^5$ per microliter has been set to diagnose PT. Tefferi *et al.*, *Mayo Clin Proc* 69:651 (1994). Most patients are asymptomatic when PT is diagnosed, usually through incidental discovery of increased peripheral blood platelet count. Bennett and Plum, *supra*, at pp. 922. Approximately one quarter, however, have either thrombotic or hemorrhagic events. *Id.* PT rarely transforms into acute leukemia or AMM, and most patients have a normal life expectancy. *Id.* at pp. 923. However, at least one third of patients with PT eventually undergo major thrombohemorrhage complications. *Id.*

In CML patients, normal bone marrow function is typically retained during the early stage. Tierney *et al*, *supra*, at pp. 503. The disease usually remains stable for years and then transforms to a more overtly malignant disease. *Id.* CML eventually progresses to blast crisis, which is indistinguishable from acute leukemia. *Id.* CML is typically a disorder of middle age (median age at presentation is 42 years). *Id.* Acceleration of the disease is often associated with fever in the absence of infection, bone pain, and splenomegaly. *Id.* One of the hallmarks of CML laboratory findings is an elevated white blood count: the median white blood count at diagnosis is 150,000/μL. *Id.* Median survival of CML is 3-4 years. *Id.* at pp. 505. Once the disease has progressed to the accelerated or blast phase, survival is typically measured in months. *Id.*

AMM is characterized by fibrosis of the bone marrow, splenomegaly, and a leukoerythroblastic peripheral blood picture with teardrop poikilocytosis. Tierney *et al*, *supra*, at pp. 502. AMM develops in adults over age 50 and is usually insidious in onset. *Id.* Later in the course of the disease, bone marrow failure takes place as the marrow becomes progressively more fibrotic. *Id.* Anemia becomes severe. *Id.* Painful episodes of splenic infarction may occur. Severe bone pain and liver failure also occur in the late stage of AMM. *Id.* The median survival from time of diagnosis is approximately 5 years. *Id.* at pp. 503.

The precise cause of MPD is not clear. Current data suggest some growth factors are involved. For instance, in both PRV and PT, in contrast to normal erythroid progenitor cells, polycythemia vera erythroid progenitor cells can grow *in vitro* in the absence of erythropoietin due to hypersensitivity to insulin like growth factor I. *Harrison's Principles of Internal Medicine*, pp. 701 (15^{th} ed., Braunwald *et al.* ed., McGraw-Hill, 2001). In AMM, the overproduction of type III collagen has been attributed to platelet-derived

growth factor or transforming growth factor β (TGF-β). *Id.* at pp. 703; see also, Martyr_, *Leuk Lymphoma* 6:1 (1991).

In some MPD forms, specific chromosomal changes are seen. For instance, nonrandom chromosome abnormalities, such as 20q-, trisomy 8 or 9 have been documented in a small percentage of untreated PRV patients, and 20q-, 13q-, trisomy 1q are common in AMM patient. *Harrison's Principles of Internal Medicine*, pp. 701-3 (15th ed., Braunwald *et al.* ed., McGraw-Hill, 2001). Philadelphia chromosome is present in the bone marrow cells of more than 90% of patients with typical CML and some patients with PRV. *See e.g.*, Kurzrock *et al.*, *N Engl J Med* 319:990 (1988). The Philadelphia chromosome results from a balanced translocation of material between the long arms of chromosomes 9 and 22. The break, which occurs at band q34 of the long arm of chromosome 9, allows translocation of the cellular oncogene C-ABL to a position on chromosome 22 called the breakpoint cluster region (bcr). The apposition of these two genetic sequences produces a new hybrid gene (BCR/ABL), which codes for a novel protein of molecular weight 210,000 kD (P210). The P210 protein, a tyrosine kinase, may play a role in triggering the uncontrolled proliferation of CML cells. *See e.g.*, Daley *et al.*, *Science* 247:824 (1990).

The risk of the CML type of MPD also increases upon exposure to ionizing radiation. Survivors of the atomic bomb explosions in Japan in 1945 have had an increased incidence of CML, with a peak occurring 5 to 12 years after exposure and seeming to be dose related. *Cecil Textbook of Medicine*, pp. 925-926 (20th ed., Bennett and Plum ed., W.B. Saunders Company, 1996). Radiation treatment of ankylosing spondylitis and cervical cancer has increased the incidence of CML. *Id*

The incidence of MPD varies depending on the form of the disease. CML constitutes one fifth of all cases of Leukemia in the United States. *Id.* at pp. 920. Approximately 4300 new cases of CML are diagnosed in the United States every year, accounting for more than half of MPD cases. (eMedicine website, myeloproliferative disease). PRV is diagnosed in 5-17 persons per 1,000,000 per year. *Id.* True incidences of PT and AMM are not known because epidemiological studies on these disorders are inadequate. *Id.* Internationally, CML appears to affect all races with approximately equal frequency. PRV is reportedly lower in Japan, *i.e.*, 2 person per 1,000,000 per year. *Id.*

2.2 MPD TREATMENT

The treatment of choice for PRV is phlebotomy. *Current Medical Diagnosis & Treatment*, pp. 501 (37th ed., Tierney *et al.* ed, Appleton & Lange, 1998). One unit of blood (approximately 500 mL) is removed weekly until the hematocrit is less than 45%. *Id.*

Because repeated phlebotomy produces iron deficiency, the requirement for phlebotomy has to be gradually decreased. *Id.* It is important to avoid medicinal iron supplementation, as this can thwart the goals of a phlebotomy program. *Id.*

In more severe cases of PRV, myelosuppressive therapy is used. *Id.* One of the widely used myelosuppressive agents is hydroxyurea. *Id.* Hydroxyurea is an oral agent that inhibits ribonucleotide reductase. Bennett and Plum, *supra*, at pp. 924. The usual dose is 500-1500 mg/d orally, adjusted to keep platelets < 500,000/μL without reducing the neurophil count to < 2000/μL. Tierney *et al.*, *supra*, at pp. 501. Side effects of hydroxyurea include mild gastrointestinal complaints, reversible neutropenia, and mucocutaneous lesions. Bennett and Plum, *supra*, at pp. 924. Busulfan may also be used in a dose of 4-6 mg/d for 4-8 weeks. Tierney *et al.*, *supra*, at pp. 501. Alpha interferon has been shown to have some ability to control the disease. The usual dose is 2-5 million units subcutaneously three times weekly. *Id.* Anagrelide has also been approved for use in treatment of thrombocytosis. *Id.* Some of the myelosuppressive agents, such as alkylating agents and radiophosphorus (^{32}P), have been shown to increase the risk of conversion of PRV to acute leukemia. *Id.* Using myelosuppressive agents for long period may cause prolonged severe myelosuppression.

Most authorities agree that treatment of PT should be aimed at decreasing the level of platelets in patients with a history of thrombosis as well as those with cardiovascular risk factors. Bennett and Plum, *supra*, at pp. 923. However, the benefit of specific therapy has not been established, and there is concern about the leukemogenic potential of the available therapeutic agents. *Id.* When treatment is decided upon, the initial drugs are hydroxyurea or anagrelide. *Id.* at pp. 924. Anagrelide is an oral agent that may involve inhibition of megakaryocyte maturation. *Id.* The starting dose is 0.5 mg given four times a day. *Id.* It is relatively contraindicated in elderly patients with heart disease. *Id.* Alpha interferon can also be used in the treatment of PT. *Id.*

Currently, there is no specific treatment for AMM. Tierney *et al.*, *supra*, at pp. 502. The management of AMM is directed to symptoms. Anemic patients are supported with red blood cells in transfusion. *Id.* Androgens such as oxymetholone, 200 mg orally daily, or testosterone help reduce the transfusion requirement in one third of cases but are poorly tolerated by women. *Id.* Splenectomy is indicated for splenic enlargement that causes recurrent painful episodes, severe thrombocytopenia, or an unacceptable high red blood cell transfusion requirement. *Id.* Alpha interferon (2-5 million units subcutaneously three times weekly) leads to improvement in some cases. *Id.*

Immediate treatment of CML is not necessary unless the white blood cell (WBC) count exceeds 200,000 per microliter or there is evidence of leukostasis (priapism, venous thrombosis, confusion, or dyspnes) or there is splenic infarction. *Id.* at pp. 504. Standard therapy of CML consists of administration of hydroxyurea. *Id.* Hydroxyurea must be given without interruption, since the white blood count will rise within days after discontinuing the medication. *Id.* Recombinant alpha interferon has largely replaced hydroxyurea as the initial treatment of choice and can prolong both the duration of the chronic phase and overall survival. *Id.* Interferon, unlike other palliative agents, can suppress the Philadelphia chromosome and to allow cytogenetically normal cells to appear. *Id.*

Although the response to myelosuppressive therapy of the chronic phase of CML is gratifying, the treatment is only palliative, and the disease is invariably fatal. *Id.* The only available curative therapy is allogenic bone marrow transplantation. *Id.* This treatment is available for adults under age 60 who have HLA-matched siblings. *Id.* Approximately 60% of adults have long term disease-free survival following bone marrow transplantation. *Id.* However, such treatment is limited by the donor source and the age of the patient. For CML patients who relapse after transplantation, immunologic therapy with infusion of T lymphocytes from the bone marrow donor may produce long-lasting remissions. *Id.* at pp 504-5. Blast crisis of CML can be treated with daunorubicin, cincristine, and prednisone (used in treatment of acute lymphoblastic leukemia), although the remission is usually short-lived. *Id.* at pp. 505.

Persistent efforts have been made to find new ways to treat CML. For instance, the synthetic inhibitor of the BCR/ABL kinase, ST1571, induces selective inhibition in the growth of t(9;22)-bearing tumor cells *in vitro* and some responses in patients. *See, e.g.*, Buchdunger *et al.*, *Proc. Natl. Acad. Sci. USA* 92:2558-2562 (1995); and Buchdunger *et al.*, *Cancer Res.*, 56:100-104 (1996). See also *Harrison's Principles of Internal Medicine*, pp. 714 (15^{th} ed., Braunwald *et al.* ed., McGraw-Hill, 2001). Inhibition of RAS with a farnesyl transferase inhibitor that blocks its insertion into the membrane may have antitumor activity in CML based on early clinical trials. *See* Braunwald *et al.*, *supra,* at 714. Preclinical efforts to use BCR/ABL peptides as a tumor vaccine appear promising. *Id.* The use of BCR/ABL antisense oligonucleotides to purge residual leukemic cells from autologous hematopoietic progenitors before reinfusion, as will as approaches to induce GVL (graft-versus-leukemia) in the setting of minimal residual disease (remission stage wherein the leukemia cell counts are below what can be detected by the traditional technology, usually

$\leq 10^{10}$ malignant cells) without inducing GVHD (graft-versus-host disease), are underway. *Id.*

Since most therapies used in the treatment of MPD are targeted only at symptoms, and most agents used have serious side effects, with the danger of causing severe myelosuppression or converting the disorder to acute leukemia, there is a great need to find new treatments of MPD that either target the underlying cause of the disorder or improve the effectiveness and safety of the current treatments.

2.3 SELECTIVE CYTOKINE INHIBITORY DRUGS

Compounds referred to as SelCIDs™ (Celgene Corporation) or Selective Cytokine Inhibitory Drugs have been synthesized and tested. These compounds potently inhibit TNF-α production, but exhibit modest inhibitory effects on LPS induced IL1ß and IL12, and do not inhibit IL6 even at high drug concentrations. In addition, SelCIDs™ tend to produce a modest IL10 stimulation. L.G. Corral, *et al.*, Ann. Rheum. Dis. 58:(Suppl I) 1107-1113 (1999).

Further characterization of the selective cytokine inhibitory drugs shows that they are potent PDE4 inhibitors. PDE4 is one of the major phosphodiesterase isoenzymes found in human myeloid and lymphoid lineage cells. The enzyme plays a crucial part in regulating cellular activity by degrading the ubiquitous second messenger cAMP and maintaining it at low intracellular levels. *Id.* Inhibition of PDE4 activity results in increased cAMP levels leading to the modulation of LPS induced cytokines including inhibition of TNF-α production in monocytes as well as in lymphocytes.

3. SUMMARY OF THE INVENTION

This invention encompasses methods of treating and preventing myeloproliferative disease ("MPD") which comprise administering to a patient in need thereof a therapeutically or prophylactically effective amount of a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof. The invention also encompasses methods of managing MPD (*e.g.*, lengthening the time of remission) which comprise administering to a patient in need of such management a therapeutically or prophylactically effective amount of a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

One embodiment of the invention encompasses the use of one or more selective cytokine inhibitory drugs in combination with conventional therapies presently used to

treat, prevent or manage MPD such as, but not limited to, hydroxyurea, anagrelide, interferons, kinase inhibitors, cancer chemotherapeutics, stem cell transplantations and other transplantations.

Another embodiment of the invention encompasses a method of reducing or preventing an adverse effect associated with MPD therapy, which comprises administering to a patient in need of such treatment or prevention an amount of a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, that is sufficient to reduce an adverse effect associated with the MPD therapy. This emodiment includes the use of a selective cytokine inhibitory drug of the invention to protect against or treat an adverse effect associated with the use of the MPD therapy. This embodiment encompasses raising a patient's tolerance for the MPD therapy.

Another embodiment of the invention encompasses a method of increasing the therapeutic efficacy of a MPD treatment which comprises administering to a patient in need of such increased therapeutic efficacy an amount of a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, that is sufficient to increase the therapeutic efficacy of the MPD treatment.

The invention further encompasses pharmaceutical compositions, single unit dosage forms, and kits suitable for use in treating, preventing and/or managing MPD, which comprise a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

4. DETAILED DESCRIPTION OF THE INVENTION

A first embodiment of the invention encompasses methods of treating or preventing MPD, which comprise administering to a patient in need of such treatment or prevention a therapeutically or prophylactically effective amount of a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof. The embodiment encompasses the treatment, prevention or management of specific sub-types of MPD such as, but not limited to, polycythemia rubra vera (PRV), primary thromobocythemia (PT), chronic myelogenous leukemia (CML), and agnogenic myeloid metaplasia (AMM).

As used herein, the term "myeloproliferative disease," or "MPD," means a hematopoietic stem cell disorder characterized by one or more of the following: clonal expansion of a multipotent hematopoietic progenitor cell with the overproduction of one or

more of the formed elements of the blood (*e.g.*, elevated red blood cell count, elevated white blood cell count, and/or elevated platelet count), presence of Philadelphia chromosome or bcr-abl gene, teardrop poikilocytosis on peripheral blood smear, leukoerythroblastic blood pictuer, giant abnormal platelets, hypercellular bone marrow with reticular or collagen fibrosis, marked left-shifted myeloid series with a low percentage of promyelocytes and blasts, splenomegaly, thrombosis, risk of progression to acute leukemia or cellular marrow with impaired morphology. The term "myeloproliferative disease," or "MPD," unless otherwise noted includes: polycythemia rubra vera (PRV), primary thromobocythemia (PT), chronic myelogenous leukemia (CML), and agnogenic myeloid metaplasia (AMM). In a specific embodiment, the term "myeloproliferative disease" or "MPD" excludes leukemia. Particular types of MPD are PRV, PT, CML and AMM.

Another embodiment of the invention encompasses methods of managing MPD which comprises administering to a patient in need of such management a prophylactically effective amount of a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

Another embodiment of the invention encompasses a pharmaceutical composition comprising a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

Also encompassed by the invention are single unit dosage forms comprising a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

Another embodiment of the invention encompasses a method of treating, preventing and/or managing MPD, which comprises administering to a patient in need of such treatment, prevention and/or management a therapeutically or prophylactically effective amount of a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, and a therapeutically or prophylactically effective amount of a second active agent.

Examples of second active agents include, but are not limited to, cytokines, corticosteroids, ribonucleotide reductase inhibitors, platelet inhibitors, all-trans retinoic acids, kinase inhibitors, topoisomerase inhibitors, farnesyl transferase inhibitors, antisense oligonucleotides, vaccines, anti-cancer agents, anti-fungal agents, anti-inflammatory agents, immunosuppressive or myelosuppressive agents, and conventional therapies for MPD.

Without being limited by theory, it is believed that certain selective cytokine inhibitory drugs can act in complementary or synergistic ways with conventional and other therapies in the treatment or management of MPD. It is also believed that certain selective

cytokine inhibitory drugs act by different mechanisms than conventional and other therapies in the treatment or management of MPD. In addition, it is believed that certain selective cytokine inhibitory drugs are effective when administered to patients who are refractory to conventional treatments for myeloproliferative diseases as well as treatments using thalidomide. As used herein, the term "refractory" means the patient's response to a MPD treatment is not satisfactory by clinical standards, *e.g.*, show no or little improvement of symptoms or laboratory findings.

It is also believed that certain therapies may reduce or eliminate particular adverse effects associated with some selective cytokine inhibitory drugs of the invention, thereby allowing the administration of larger amounts of a selective cytokine inhibitory drug to patients and/or increasing patient compliance. It is further believed that some selective cytokine inhibitory drugs may reduce or eliminate particular adverse effects associated with other MPD therapies, thereby allowing the administration of larger amounts of such therapies to patients and/or increasing patient compliance.

Another embodiment of the invention encompasses a kit comprising: a pharmaceutical composition comprising a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof and a second active agent and/or instructions for use. The invention further encompasses kits comprising single unit dosage forms.

Another embodiment of the invention encompasses a method of reversing, reducing or avoiding an adverse effect associated with the administration of an active agent used to treat MPD in a patient suffering from MPD, which comprises administering to a patient in need thereof a therapeutically or prophylactically effective amount of a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof. Examples of active agents include, but are not limited to, the second active agents described herein (*see* section 4.2.).

Examples of adverse effects associated with active agents used to treat MPD include, but are not limited to: conversion to acute leukemia; severe myelosuppression; gastrointestinal toxicity such as, but not limited to, early and late-forming diarrhea and flatulence; gastrointestinal bleeding; nausea; vomiting; anorexia; leukopenia; anemia; neutropenia; asthenia; abdominal cramping; fever; pain; loss of body weight; dehydration; alopecia; dyspnea; insomnia; dizziness, mucositis, xerostomia, mucocutaneous lesions, and kidney failure.

As leukemic transformation develops in certain stages of MPD, transplantation of peripheral blood stem cells, hematopoietic stem cell preparation or bone marrow may be necessary. Without being limited by theory, it is believed that the combined use of a selective cytokine inhibitory drug and the transplantation of stem cells in a patient suffering from MPD provides a unique and unexpected synergism. In particular, it is believed that a selective cytokine inhibitory drug exhibits immunomodulatory activity that can provide additive or synergistic effects when given concurrently with transplantation therapy. Selective cytokine inhibitory drugs of the invention can work in combination with transplantation therapy to reduce complications associated with the invasive procedure of transplantation and risk of related Graft Versus Host Disease (GVHD). Therefore, this invention encompasses a method of treating, preventing and/or managing MPD, which comprises administering to a patient (*e.g.*, a human) a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, before, during, or after transplantation therapy.

The invention also encompasses pharmaceutical compositions, single unit dosage forms, and kits which comprise one or more selective cytokine inhibitory drugs of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, a second active ingredient, and/or blood or cells for transplantation therapy. For example, a kit may contain one or more compounds of the invention, stem cells for transplantation and an immunosuppressive agent, and an antibiotic or other drug.

4.1 SELECTIVE CYTOKINE INHIBITORY DRUGS

Compounds used in the invention include racemic, stereomerically pure or stereomerically enriched selective cytokine inhibitory drugs, stereomerically or enantiomerically pure compounds that have selective cytokine inhibitory activities, and pharmaceutically acceptable salts, solvates, hydrates, stereoisomers, clathrates, and prodrugs thereof. Preferred compounds used in the invention are known Selective Cytokine Inhibitory Drugs (SelCIDs™) of Celgene Corporation.

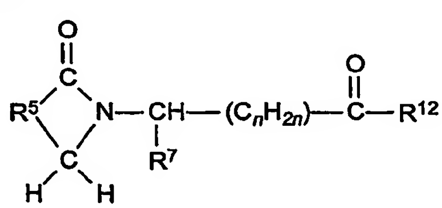
As used herein and unless otherwise indicated, the term "SelCIDs™" used in the invention encompasses small molecule drugs, *e.g.*, small organic molecules which are not peptides, proteins, nucleic acids, oligosaccharides or other macromolecules. Preferred compounds inhibit TNF-α production. Further, the compounds may also have a modest inhibitory effect on LPS induced IL1ß and IL12. More preferably, the compounds of the invention are potent PDE4 inhibitors. PDE4 is one of the major phosphodiesterase isoenzymes found in human myeloid and lymphoid lineage cells. The enzyme plays a crucial part in regulating cellular activity by degrading the ubiquitous second messenger

cAMP and maintaining it at low intracellular levels. Without being limited by theory, inhibition of PDE4 activity results in increased cAMP levels leading to the modulation of LPS induced cytokines, including inhibition of TNF-α production in monocytes as well as in lymphocytes.

Specific examples of selective cytokine inhibitory drugs include, but are not limited to, the cyclic imides disclosed in U.S. patent no. 5,605,914; the cycloalkyl amides and cycloalkyl nitriles of U.S. patent nos. 5,728,844 and 5,728,845, respectively; the aryl amides (for example, an embodiment being N-benzoyl-3-amino-3-(3',4'-dimethoxyphenyl)-propanamide) of U.S. patent nos. 5,801,195 and 5,736,570; the imide/amide ethers and alcohols (for example 3-phthalimido-3-(3',4'-dimethoxypheryl)propan-1-ol) disclosed in U.S. patent no. 5,703,098; the succinimides and maleimides (for example methyl 3-(3',4',5'6'-petrahydrophthalimdo)-3-(3",4"-dimethoxyphenyl)propionate) disclosed in U.S. patent no. 5,658,940; imido and amido substituted alkanohydroxamic acids disclosed in WO 99/06041 and substituted phenethylsulfones disclosed in U.S. patent no. 6,020,358; and aryl amides such as N-benzoyl-3-amino-3-(3',4'-dimethoxyphenyl)propanamide as described in U.S. patent no. 6,046,221. The entireties of each of the patents and patent applications identified herein are incorporated herein by reference. Selective cytokine inhibitory drugs of the invention do not include thalidomide.

Additional selective cytokine inhibitory drugs belong to a family of synthesized chemical compounds of which typical embodiments include 3-(1,3-dioxobenzo-[f]isoindol-2-yl)-3-(3-cyclopentyloxy-4-methoxyphenyl)propionamide and 3-(1,3-dioxo-4-azaisoindol-2-yl)-3-(3,4-dimethoxyphenyl)-propionamide.

Other specific selective cytokine inhibitory drugs belong to a class of non-polypeptide cyclic amides disclosed in U.S. patent nos. 5,698,579 and 5,877,200, both of which are incorporated herein. Representative cyclic amides include compounds of the formula:



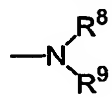
wherein n has a value of 1, 2, or 3;

R^5 is o-phenylene, unsubstituted or substituted with 1 to 4 substituents each selected independently from the group consisting of nitro, cyano, trifluoromethyl, carbethoxy, carbomethoxy, carbopropoxy, acetyl, carbamoyl, acetoxy, carboxy, hydroxy, amino,

alkylamino, dialkylamino, acylamino, alkyl of 1 to 10 carbon atoms, alkyl of 1 to 10 carbon atoms, and halo;

R^7 is (i) phenyl or phenyl substituted with one or more substituents each selected independently of the other from the group consisting of nitro, cyano, trifluoromethyl, carbethoxy, carbomethoxy, carbopropoxy, acetyl, carbamoyl, acetoxy, carboxy, hydroxy, amino, alkyl of 1 to 10 carbon atoms, alkoxy of 1 to 10 carbon atoms, and halo, (ii) benzyl unsubstituted or substituted with 1 to 3 substituents selected from the group consisting of nitro, cyano, trifluoromethyl, carbothoxy, carbomethoxy, carbopropoxy, acetyl, carbamoyl, acetoxy, carboxy, hydroxy, amino, alkyl of 1 to 10 carbon atoms, alkoxy of 1 to 10 carbon atoms, and halo, (iii) naphthyl, and (iv) benzyloxy;

R^{12} is -OH, alkoxy of 1 to 12 carbon atoms, or



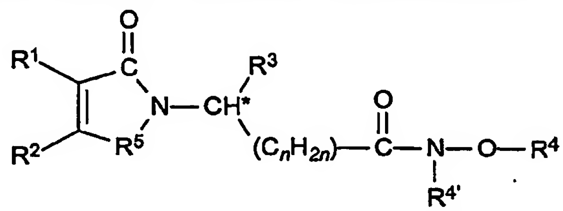
R^8 is hydrogen or alkyl of 1 to 10 carbon atoms; and

R^9 is hydrogen, alkyl of 1 to 10 carbon atoms, $-COR^{10}$, or $-SO_2R^{10}$, wherein R^{10} is hydrogen, alkyl of 1 to 10 carbon atoms, or phenyl.

Specific compounds of this class include, but are not limited to:

3-phenyl-2-(1-oxoisoindolin-2-yl)propionic acid;
3-phenyl-2-(1-oxoisoindolin-2-yl)propionamide;
3-phenyl-3-(1-oxoisoindolin-2-yl)propionic acid;
3-phenyl-3-(1-oxoisoindolin-2-yl)propionamide;
3-(4-methoxyphenyl)-3-(1-oxisoindolin-yl)propionic acid;
3-(4-methoxyphenyl)-3-(1-oxisoindolin-yl)propionamide;
3-(3,4-dimethoxyphenyl)-3-(1-oxisoindolin-2-yl)propionic acid;
3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydroisoindol-2-yl)-propionamide;
3-(3,4-dimethoxyphenyl)-3-(1-oxisoindolin-2-yl)propionamide;
3-(3,4-diethoxyphenyl)-3-(1-oxoisoindolin-yl)propionic acid;
methyl 3-(1-oxoisoindolin-2-yl)-3-(3-ethoxy-4-methoxyphenyl)propionate;
3-(1-oxoisoindolin-2-yl)-3-(3-ethoxy-4-methoxyphenyl)propionic acid;
3-(1-oxoisoindolin-2-yl)-3-(3-propoxy-4-methoxyphenyl)propionic acid;
3-(1-oxoisoindolin-2-yl)-3-(3-butoxy-4-methoxyphenyl)propionic acid;
3-(1-oxoisoindolin-2-yl)-3-(3-propoxy-4-methoxyphenyl)propionamide;
3-(1-oxoisoindolin-2-yl)-3-(3-butoxy-4-methoxyphenyl)propionamide;
methyl 3-(1-oxoisoindolin-2-yl)-3-(3-butoxy-4-methoxyphenyl)propionate; and
methyl 3-(1-oxoisoindolin-2-yl)-3-(3-propoxy-4-methoxyphenyl)propionate.

Other specific selective cytokine inhibitory drugs include the imido and amido substituted alkanohydroxamic acids disclosed in WO 99/06041, which is incorporated herein by reference. Examples of such compound include, but are not limited to:



wherein each of R^1 and R^2, when taken independently of each other, is hydrogen, lower alkyl, or R^1 and R^2, when taken together with the depicted carbon atoms to which each is bound, is *o*-phenylene, *o*-naphthylene, or cyclohexene-1,2-diyl, unsubstituted or substituted with 1 to 4 substituents each selected independently from the group consisting of nitro, cyano, trifluoromethyl, carbethoxy, carbomethoxy, carbopropoxy, acetyl, carbamoyl, acetoxy, carboxy, hydroxy, amino, alkylamino, dialkylamino, acylamino, alkyl of 1 to 10 carbon atoms, alkoxy of 1 to 10 carbon atoms, and halo;

R^3 is phenyl substituted with from one to four substituents selected from the group consisting of nitro, cyano, trifluoromethyl, carbethoxy, carbomethoxy, carbopropoxy, acetyl, carbamoyl, acetoxy, carboxy, hydroxy, amino, alkyl of 1 to 10 carbon atoms, alkoxy of 1 to 10 carbon atoms, alkylthio of 1 to 10 carbon atoms, benzyloxy, cycloalkoxy of 3 to 6 carbon atoms, C_4-C_6-cycloalkylidenemethyl, C_3-C_{10}-alkylidenemethyl, indanyloxy, and halo;

R^4 is hydrogen, alkyl of 1 to 6 carbon atoms, phenyl, or benzyl;

$R^{4'}$ is hydrogen or alkyl of 1 to 6 carbon atoms;

R^5 is $-CH_2-$, $-CH_2-CO-$,$-SO_2-$,-S-, or -NHCO-;

n has a value of 0, 1, or 2; and

the acid addition salts of said compounds which contain a nitrogen atom capable of being protonated.

Additional specific selective cytokine inhibitory drugs used in the invention include, but are not limited to:

3-(3-ethoxy-4-methoxyphenyl)-N-hydroxy-3-(1-oxoisoindolinyl)propionamide;

3-(3-ethoxy-4-methoxyphenyl)-N-methoxy-3-(1-oxoisoindolinyl)propionamide;

N-benzyloxy-3-(3-ethoxy-4-methoxyphenyl)-3-phthalimidopropionamide;

N-benzyloxy-3-(3-ethoxy-4-methoxyphenyl)-3-(3-nitrophthalimido)propionamide;

N-benzyloxy-3-(3-ethoxy-4-methoxyphenyl)-3-(1-oxoisoindolinyl)propionamide;

3-(3-ethoxy-4-methoxyphenyl)-N-hydroxy-3-phthalimidopropionamide;

N-hydroxy-3-(3,4-dimethoxyphenyl)-3-phthalimidopropionamide;

3-(3-ethoxy-4-methoxyphenyl)-N-hydroxy-3-(3-nitrophthalimido)propionamide;

N-hydroxy-3-(3,4-dimethoxyphenyl)-3-(1-oxoisoindolinyl)propionamide;

3-(3-ethoxy-4-methoxyphenyl)-N-hydroxy-3-(4-methyl-phthalimido)propionamide;

3-(3-cyclopentyloxy-4-methoxyphenyl)-N-hydroxy-3-phthalimidopropionamide;

3-(3-ethoxy-4-methoxyphenyl)-N-hydroxy-3-(1,3-dioxo-2,3-dihydro-1H-benzo[f]isoindol-2-yl)propionamide;

N-hydroxy-3-{3-(2-propoxy)-4-methoxyphenyl}-3-phthalimidopropionamide;

3-(3-ethoxy-4-methoxyphenyl)-3-(3,6-difluorophthalimido)-N-hydroxypropionamide;

3-(4-aminophthalimido)-3-(3-ethoxy-4-methoxyphenyl)-N-hydroxypropionamide;

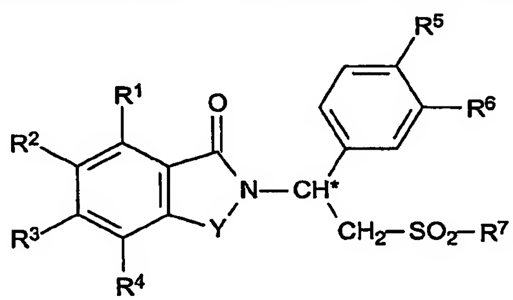
3-(3-aminophthalimido)-3-(3-ethoxy-4-methoxyphenyl)-N-hydroxypropionamide;

N-hydroxy-3-(3,4-dimethoxyphenyl)-3-(1-oxoisoindolinyl)propionamide;

3-(3-cyclopentyloxy-4-methoxyphenyl)-N-hydroxy-3-(1-oxoisoindolinyl) propionamide; and

N-benzyloxy-3-(3-ethoxy-4-methoxyphenyl)-3-(3-nitrophthalimido)propionamide.

Additional selective cytokine inhibitory drugs used in the invention include the substituted phenethylsulfones substituted on the phenyl group with a oxoisoindine group. Examples of such compounds include, but are not limited to, those disclosed in U.S. patent no. 6,020,358, which is incorporated herein, which include the following:



wherein the carbon atom designated * constitutes a center of chirality;

Y is C=O, CH2, SO_2, or CH_2C=O; each of R^1, R^2, R^3, and R^4, independently of the others, is hydrogen, halo, alkyl of 1 to 4 carbon atoms, alkoxy of 1 to 4 carbon atoms, nitro, cyano, hydroxy, or $-NR^8R^9$; or any two of R^1, R^2, R^3, and R^4 on adjacent carbon atoms, together with the depicted phenylene ring are naphthylidene;

each of R^5 and R^6, independently of the other, is hydrogen, alkyl of 1 to 4 carbon atoms,alkoxy of 1 to 4 carbon atoms, cyano, or cycloalkoxy of up to 18 carbon atoms;

R^7 is hydroxy, alkyl of 1 to 8 carbon atoms, phenyl, benzyl, or $NR^{8'}R^{9'}$;

each of R^8 and R^9 taken independently of the other is hydrogen, alkyl of 1 to 8 carbon atoms, phenyl, or benzyl, or one of R^8 and R^9 is hydrogen and the other is $-COR^{10}$ or

$-SO_2R^{10}$, or R^8 and R^9 taken together are tetramethylene, pentamethylene, hexamethylene, or $-CH_2CH_2X^1CH_2CH_2-$ in which X^1 is -O-, -S- or -NH-; and

each of $R^{8'}$ and $R^{9'}$ taken independently of the other is hydrogen, alkyl of 1 to 8 carbon atoms, phenyl, or benzyl, or one of $R^{8'}$ and $R^{9'}$ is hydrogen and the other is $-COR^{10'}$ or $-SO_2R^{10'}$, or $R^{8'}$ and $R^{9'}$ taken together are tetramethylene, pentamethylene, hexamethylene, or $-CH_2CH_2X^2CH_2CH_2-$ in which X^2 is -O-, -S-, or -NH-.

It will be appreciated that while for convenience the above compounds are identified as phenethylsulfones, they include sulfonamides when R^7 is $NR^{8'}R^{9'}$.

Specific groups of such compounds are those in which Y is C=O or CH_2.

A further specific group of such compounds are those in which each of R^1, R^2, R^3, and R^4 independently of the others, is hydrogen, halo, methyl, ethyl, methoxy, ethoxy, nitro, cyano, hydroxy, or $-NR^8R^9$ in which each of R^8 and R^9 taken independently of the other is hydrogen or methyl or one of R^8 and R^9 is hydrogen and the other is $-COCH_3$.

Particular compounds are those in which one of R^1, R^2, R^3, and R^4 is $-NH_2$ and the remaining of R^1, R^2, R^3, and R^4 are hydrogen.

Particular compounds are those in which one of R^1, R^2, R^3, and R^4 is $-NHCOCH_3$ and the remaining of R^1, R^2, R^3, and R^4 are hydrogen.

Particular compounds are those in which one of R^1, R^2, R^3, and R^4 is $-N(CH_3)_2$ and the remaining of R^1, R^2, R^3, and R^4 are hydrogen.

A further preferred group of such compounds are those in which one of R^1, R^2, R^3, and R^4 is methyl and the remaining of R^1, R^2, R^3, and R^4 are hydrogen.

Particular compounds are those in which one of R^1, R^2, R^3, and R^4 is fluoro and the remaining of R^1, R^2, R^3, and R^4 are hydrogen.

Particular compounds are those in which each of R^5 and R^6, independently of the other, is hydrogen, methyl, ethyl, propyl, methoxy, ethoxy, propoxy, cyclopentoxy, or cyclohexoxy.

Particular compounds are those in which R^5 is methoxy and R^6 is monocycloalkoxy, polycycloalkoxy, and benzocycloalkoxy.

Particular compounds are those in which R^5 is methoxy and R^6 is ethoxy.

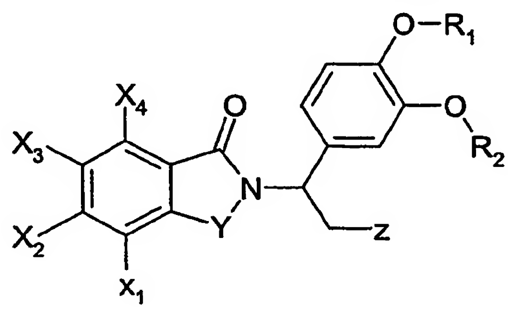
Particular compounds are those in which R^7 is hydroxy, methyl, ethyl, phenyl, benzyl, or $NR^{8'}R^{9'}$ in which each of $R^{8'}$ and $R^{9'}$ taken independently of the other is hydrogen or methyl.

Particular compounds are those in which R^7 is methyl, ethyl, phenyl, benzyl or $NR^{8'}R^{9'}$ in which each of $R^{8'}$ and $R^{9'}$ taken independently of the other is hydrogen or methyl.

Particular compounds are those in which R^7 is methyl.

Particular compounds are those in which R^7 is $NR^{8'}R^{9'}$ in which each of $R^{8'}$ and $R^{9'}$ taken independently of the other is hydrogen or methyl.

Other specific selective cytokine inhibitory drugs include fluoroalkoxy-substituted 1,3-dihydro-isoindolyl compounds found in United States Provisional Application No. 60/436,975 to G. Muller *et al.*, filed December 30, 2002, which is incorporated herein in its entirety by reference. Representative fluoroalkoxy-substituted 1,3-dihydro-isoindolyl compounds include compounds of the formula:



wherein:

Y is -C(O)-, $-CH_2$, $-CH_2C(O)-$, $-C(O)CH_2-$, or SO_2;

Z is –H, $-C(O)R^3$, $-(C_{0-1}\text{-alkyl})-SO_2-(C_{1-4}\text{-alkyl})$, $-C_{1-8}$-alkyl, $-CH_2OH$, $CH_2(O)(C_{1-8}\text{-alkyl})$ or -CN;

R_1 and R_2 are each independently $-CHF_2$, $-C_{1-8}$-alkyl, $-C_{3-18}$-cycloalkyl, or $-(C_{1-10}\text{-alkyl})(C_{3-18}\text{-cycloalkyl})$, and at least one of R_1 and R_2 is CHF_2;

R^3 is $-NR^4R^5$, -alkyl, -OH, –O-alkyl, phenyl, benzyl, substituted phenyl, or substituted benzyl;

R^4 and R^5 are each independently –H, $-C_{1-8}$-alkyl, -OH, $-OC(O)R^6$;

R^6 is $-C_{1-8}$-alkyl, -amino(C_{1-8}-alkyl), -phenyl, -benzyl, or -aryl;

X_1, X_2, X_3, and X_4 are each independent -H, -halogen, -nitro, $-NH_2$, $-CF_3$, $-C_{1-6}$-alkyl, $-(C_{0-4}\text{-alkyl})-(C_{3-6}\text{-cycloalkyl})$, $(C_{0-4}\text{-alkyl})-NR^7R^8$, $(C_{0-4}\text{-alkyl})-N(H)C(O)-(R^8)$, $(C_{0-4}\text{-alkyl})-N(H)C(O)N(R^7R^8)$, $(C_{0-4}\text{-alkyl})-N(H)C(O)O(R^7R^8)$, $(C_{0-4}\text{-alkyl})-OR^8$, $(C_{0-4}$-alkyl)-imidazolyl, $(C_{0-4}$-alkyl)-pyrrolyl, $(C_{0-4}$-alkyl)-oxadiazolyl, or $(C_{0-4}$-alkyl)-triazolyl, or two of X_1, X_2, X_3, and X_4 may be joined together to form a cycloalkyl or heterocycloalkyl ring, (*e.g.*, X_1 and X_2, X_2 and X_3, X_3 and X_4, X_1 and X_3, X_2 and X_4, or X_1 and X_4 may form a 3, 4, 5, 6, or 7 membered ring which may be aromatic, thereby forming a bicyclic system with the isoindolyl ring); and

R^7 and R^8 are each independently H, C_{1-9}-alkyl, C_{3-6}-cycloalkyl, $(C_{1-6}\text{-alkyl})-(C_{3-6}\text{-cycloalkyl})$, $(C_{1-6}\text{-alkyl})-N(R^7R^8)$, $(C_{1-6}\text{-alkyl})-OR^8$, phenyl, benzyl, or aryl;

or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

Preferred compounds include, but are not limited to:

3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(3-cyclopropylmethoxy-4-difluoromethoxy-phenyl)-propionic acid;

3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(3-cyclopropylmethoxy-4-difluoromethoxy-phenyl)-N,N-dimethyl-propionamide;

3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(3 cyclopropylmethoxy-4-difluoromethoxy-phenyl)-propionamide;

3-(3-Cyclopropylmethoxy-4-difluoromethoxy-phenyl)-3-(1,3-dioxo-1,3-dihydro-isoindol-2-yl)-propionic acid;

3-(3-Cyclopropylmethoxy-4-difluoromethoxy-phenyl)-3-(1,3-dioxo-1,3-dihydro-isoindol-2-yl)-N-hydroxy-propionamide;

3-(3-Cyclopropylmethoxy-4-difluoromethoxy-phenyl)-3-(7-nitro-1-oxo-1,3-dihydro-isoindol-2-yl)-propionic acid methyl ester;

3-(3-Cyclopropylmethoxy-4-difluoromethoxy-phenyl)-3-(7-nitro-1-oxo-1,3-dihydro-isoindol-2-yl)-propionic acid;

3-(3-Cyclopropylmethoxy-4-difluoromethoxy-phenyl -3-(7-nitro-1-oxo-1,3-dihydro-isoindol-2-yl)-)-N,N-dimethyl-propionamide;

3-(7-Amino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(3-cyclopropylmethoxy-4-difluoromethoxy-phenyl)-N,N-dimethyl-propionamide;

3-(4-Difluoromethoxy-3-ethoxy-phenyl)-3-(7-nitro-1-oxo-1,3-dihydro-isoindol-2-yl)-propionic acid methyl ester;

3-(7-Amino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionic acid methyl ester;

3-[7-(Cyclopropanecarbonyl-amino)-1-oxo-1,3-dihydro-isoindol-2-yl]-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionic acid methyl ester;

3-(7-Acetylamino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionic acid methyl ester;

3-(7-Acetylamino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionic acid;

3-[7-(Cyclopropanecarbonyl-amino)-1-oxo-1,3-dihydro-isoindol-2-yl]-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionic acid;

Cyclopropanecarboxylic acid {2-[2-carbamoyl-1-(4-difluoromethoxy-3-ethoxy-phenyl)-ethyl]-3-oxo-2,3-dihydro-1H-isoindol-4-yl}-amide;

Cyclopropanecarboxylic acid {2-[1-(4-difluoromethoxy-3-ethoxy-phenyl)-2-dimethylcarbamoyl-ethyl]-3-oxo-2,3-dihydro-1H-isoindol-4-yl}-;

Cyclopropanecarboxylic acid {2-[1-(4-difluoromethoxy-3-ethoxy-phenyl)-2-hydroxycarbamoyl-ethyl]-3-oxo-2,3-dihydro-1H-isoindol-4-yl}-amide;

3-(7-Acetylamino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionamide;

3-(7-Acetylamino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-N,N-dimethyl-propionamide;

3-(7-Acetylamino-1-oxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-N-hydroxy-propionamide;

3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionic acid;

3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-propionamide;

3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-N,N-dimethyl-propionamide;

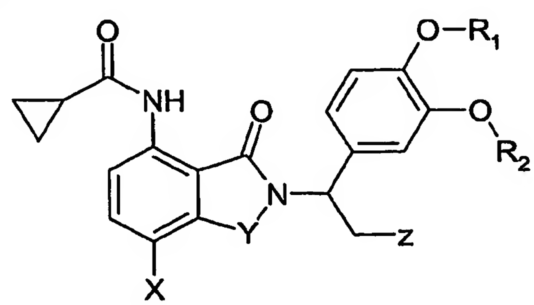
3-(4-Acetylamino-1,3-dioxo-1,3-dihydro-isoindol-2-yl)-3-(4-difluoromethoxy-3-ethoxy-phenyl)-N-hydroxy-propionamide;

Cyclopropanecarboxylic acid {2-[1-(4-difluoromethoxy-3-ethoxy-phenyl)-2-methanesulfonyl-ethyl]-3-oxo-2,3-dihydro-1H-isoindol-4-yl}-amide;

N-{2-[1-(4-Difluoromethoxy-3-ethoxy-phenyl)-2-methanesulfonyl-ethyl]-1,3-dioxo-2,3-dihydro-1H-isoindol-4-yl}-acetamide; and

Cyclopropanecarboxylic acid {2-[2-carbamoyl-1-(4-difluoromethoxy-3-ethoxy-phenyl)-ethyl]-7-chloro-3-oxo-2,3-dihydro-1H-isoindol-4-yl}-amide.

Other selective cytokine inhibitory drugs include 7-amido-substituted isoindolyl compounds found in United States Provisional Application No. 60/454,155 to G. Muller *et al.*, filed March 12, 2003, which is incorporated herein in its entirety by reference. Representative 7-amido-substituted isoindolyl compounds include compounds of the formula:



wherein:

Y is -C(O)-, $-CH_2$, $-CH_2C(O)$-or SO_2;

X is H;

Z is (C_{0-4}-alkyl)-C(O)R^3, C_{1-4}-alkyl, (C_{0-4}-alkyl)-OH, (C_{1-4}-alkyl)-O(C_{1-4}-alkyl), (C_{1-4}-alkyl)-SO_2(C_{1-4}-alkyl), (C_{0-4}-alkyl)-SO(C_{1-4}-alkyl), (C_{0-4}-alkyl)-NH_2, (C_{0-4}- alkyl)-N(C_{1-8}-alkyl)$_2$, (C_{0-4}-alkyl)-N(H)(OH), CH_2NSO_2(C_{1-4}-alkyl);

R_1 and R_2 are independently C_{1-8}-alkyl, cycloalkyl, or(C_{1-4}-alkyl)cycloalkyl;

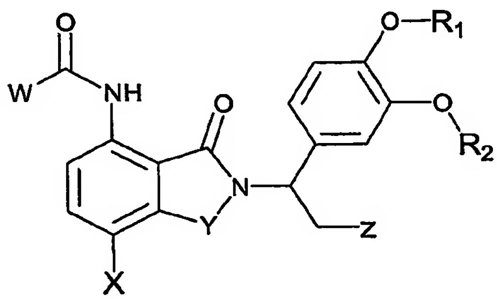
R^3 is, NR^4R^5, OH, or O-(C_{1-8}-alkyl);

R^4 is H;

R^5 is -OH, or -OC(O)R^6;

R^6 is C_{1-8}-alkyl, amino-(C_{1-8}-alkyl), (C_{1-8}-alkyl)-(C_{3-6}-cycloalkyl), C_{3-6}cycloalkyl, phenyl, benzyl, or aryl;

or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof; or the formula:

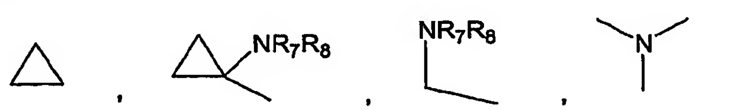


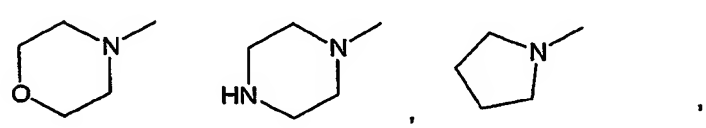
wherein:

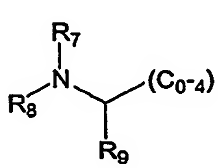
Y is -C(O)-, $-CH_2$, $-CH_2C(O)$-, or SO_2;

X is halogen, -CN, $-NR_7R_8$, $-NO_2$, or $-CF_3$,

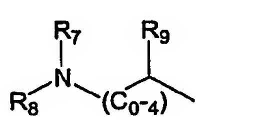
W is







or



;

Z is ($C_{0\text{-}4}$alkyl)-SO_2($C_{1\text{-}4}$-alkyl), -($C_{0\text{-}4}$alkyl)-CN, -($C_{0\text{-}4}$alkyl)-C(O)R^3, $C_{1\text{-}4}$-alkyl, ($C_{0\text{-}4}$-alkyl)OH, ($C_{0\text{-}4}$-alkyl)O($C_{1\text{-}4}$-alkyl), ($C_{0\text{-}4}$-alkyl)SO($C_{1\text{-}4}$-alkyl), ($C_{0\text{-}4}$-alkyl)NH_2, ($C_{0\text{-}4}$-alkyl)N($C_{1\text{-}8}$-alkyl)$_2$, ($C_{0\text{-}4}$-alkyl) N(H)(OH), or ($C_{0\text{-}4}$-alkyl)NSO_2(C1-4-alkyl);

W is -$C_{3\text{-}6}$-cycloalkyl, -($C_{1\text{-}8}$-alkyl)-($C_{3\text{-}6}$-cycloalkyl), -($C_{0\text{-}8}$-alkyl)-($C_{3\text{-}6}$cycloalkyl)-NR_7R_8, ($C_{0\text{-}8}$-alkyl)-NR_7R_8, ($C_{0\text{-}4}$-alkyl)-CHR_9-($C_{0\text{-}4}$-alkyl)-NR_7R_8;

R_1 and R_2 are independently $C_{1\text{-}8}$-alkyl, cycloalkyl, or ($C_{1\text{-}4}$-alkyl)cycloalkyl;

R^3 is $C_{1\text{-}8}$-alkyl, NR^4R^5, OH, or O-($C_{1\text{-}8}$-alkyl);

R^4 and R^5 are independently H, $C_{1\text{-}8}$-alkyl, ($C_{0\text{-}8}$-alkyl)-($C_{3\text{-}6}$-cycloalkyl), OH, or –OC(O)R^6;

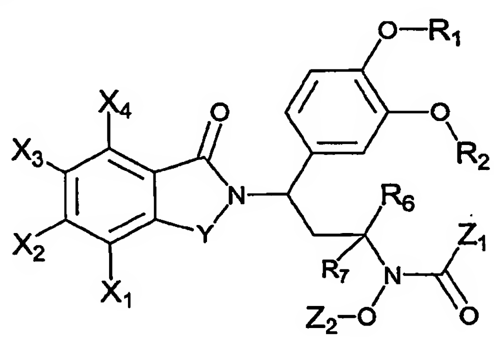
R^6 is $C_{1\text{-}8}$-alkyl, ($C_{0\text{-}8}$-alkyl)-($C_{3\text{-}6}$-cycloalkyl), amino-($C_{1\text{-}8}$-alkyl), phenyl, benzyl, or aryl;

R_7 and R_8 are each independently H, $C_{1\text{-}8}$-alkyl, ($C_{0\text{-}8}$alkyl)-($C_{3\text{-}6}$-cycloalkyl), phenyl, benzyl, aryl, or can be taken together with the atom connecting them to form a 3 to 7 membered heterocycloalkyl or heteroaryl ring;

R_9 is $C_{1\text{-}4}$-alkyl, ($C_{0\text{-}4}$-alkyl)aryl, ($C_{0\text{-}4}$-alkyl)-($C_{3\text{-}6}$-cycloalkyl), ($C_{0\text{-}4}$-alkyl)-heterocylcle;

or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

Still other selective cytokine inhibitory drugs include N-alkyl-hydroxamic acid-isoindolyl compounds found in United States Provisional Application No. 60/454,149 to G. Muller *et al.*, filed March 12, 2003, which is incorporated herein in its entirety by reference. Representative N-alkyl-hydroxamic acid-isoindolyl compounds include compounds of the formula:



wherein:

Y is -C(O)-, -CH_2, -CH_2C(O)- or SO_2;

R_1 and R_2 are independently $C_{1\text{-}8}$-alkyl, CF_2H, CF_3, CH_2CHF_2, cycloalkyl, or ($C_{1\text{-}8}$-alkyl)cycloalkyl;

Z_1 is H, $C_{1\text{-}6}$-alkyl, -NH_2 –NR_3R_4 or OR_5;

Z_2 is H or C(O)R_5;

X_1, X_2, X_3 and X_4 are each independent H, halogen, NO_2, OR_3, CF_3, C_{1-6}-alkyl, (C_{0-4}-alkyl)-(C_{3-6}-cycloalkyl), (C_{0-4}-alkyl)-N-(R_8R_9), (C_{0-4}-alkyl)-NHC(O)-(R_8), (C_{0-4}-alkyl)-NHC(O)CH(R_8)(R_9), (C_{0-4}-alkyl)-NHC(O)N(R_8R_9), (C_{0-4}-alkyl)-NHC(O)O(R_8), (C_{0-4}-alkyl)-O-R_8, (C_{0-4}-alkyl)-imidazolyl, (C_{0-4}-alkyl)-pyrrolyl, (C_{0-4}-alkyl)-oxadiazolyl, (C_{0-4}-alkyl)-triazolyl or (C_{0-4}-alkyl)-heterocycle;

R_3, R_4, and R_5 are each independently H, C_{1-6}-alkyl, O-C_{1-6}-alkyl, phenyl, benzyl, or aryl;

R_6 and R_7 are independently H or C_{1-6}-alkyl;

R_8 and R_9 are each independently H, C_{1-9}-alkyl, C_{3-6}-cycloalkyl, (C_{1-6}-alkyl)-(C_{3-6}-cycloalkyl), (C_{0-6}-alkyl)-N(R_4R_5), (C_{1-6}-alkyl)-OR_5, phenyl, benzyl, aryl, piperidinyl, piperizinyl, pyrolidinyl, morpholino, or C_{3-7}-heterocycloalkyl; or

a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof.

Specific selective cytokine inhibitory drugs include, but are not limited to:

2-[1(-3-ethoxy-4-methoxyphenyl)-2-methyl-sulfonylethyl]isoindolin-1-one;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-(N,N-dimethyl-aminosulfonyl)ethyl]isoindolin-1-one;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methyl-sulfonylethyl]isoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methyl-sulfonylethyl]-5-nitro-isoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methyl-sulfonylethyl]-4-nitroisoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-4-aminoisoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-5-methylisoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-5-acetamidoisoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-4-dimethylaminoisondoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-5-dimethylaminoisoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]benzo[e]isoindoline-1,3-dione;

2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-4-methoxyisoindoline-1,3-dione;

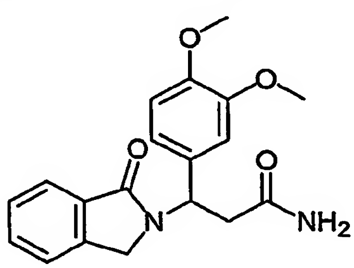
1-(3-cyclopentyloxy-4-methoxyphenyl)-2-methylsulfonylethyl-amine;

2-[1-(3-cyclopentyloxy-4-methoxyphenyl)-2-methylsulfonylethyl]isoindoline-1,3-dione; and

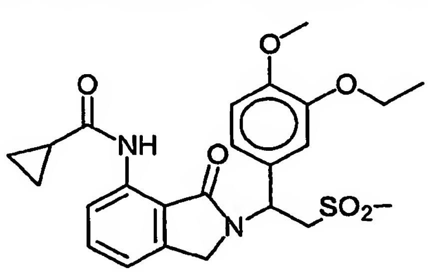
2-[1-(3-cyclopentyloxy-4-methoxyphenyl)-2-methylsulfonylethyl]-4-dimethylaminoisoindoline-1,3-dione.

Additional selective cytokine inhibitory drugs include the enantiomerically pure compounds disclosed in U.S. provisional patent application nos. 60/366,515 and 60/366,516 to G. Muller *et al.*, both of which were filed March 20, 2002; U.S. provisional patent application nos 60/438, 450 and 60/438,448 to G. Muller *et al.*, both of which were filed on Januray 7, 2003; and U.S. provisional patent application no. 60/452,460 to G. Muller *et al.* filed on March 5, 2003, all of which are incorporated herein by reference. Preferred compounds include an enantiomer of 2-[1-(3-ethoxy-4-methoxyphenyl)-2-methylsulfonylethyl]-4-acetylaminoisoindoline-1,3-dione and an enantiomer of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide.

Preferred selective cytokine inhibitory drugs used in the invention are 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide and cyclopropanecarboxylic acid {2-[1-(3-ethoxy-4-methoxy-phenyl)-2-methanesulfonyl-ethyl]-3-oxo-2,3-dihydro-1 *H*-isoindol-4-yl}-amide, which are available from Celgene Corp., Warren, NJ. 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide has the following chemical structure:



Cyclopropanecarboxylic acid {2-[1-(3-ethoxy-4-methoxy-phenyl)-methanesulfonyl-ethyl]-3-oxo-2,3-dihydro-1 *H*-isoindol-4-yl}-amide has the following chemical structure:



The compounds of the invention can either be commercially purchased or prepared according to the methods described in the patents or patent publications disclosed herein. Further, optically pure compositions can be asymmetrically synthesized or resolved using known resolving agents or chiral columns as well as other standard synthetic organic chemistry techniques.

As used herein and unless otherwise indicated, the term "pharmaceutically acceptable salt" encompasses non-toxic acid and base addition salts of the compound to which the term refers. Acceptable non-toxic acid addition salts include those derived from organic and inorganic acids or bases know in the art, which include, for example, hydrochloric acid, hydrobromic acid, phosphoric acid, sulfuric acid, methanesulphonic acid, acetic acid, tartaric acid, lactic acid, succinic acid, citric acid, malic acid, maleic acid, sorbic acid, aconitic acid, salicylic acid, phthalic acid, embolic acid, enanthic acid, and the like.

Compounds that are acidic in nature are capable of forming salts with various pharmaceutically acceptable bases. The bases that can be used to prepare pharmaceutically acceptable base addition salts of such acidic compounds are those that form non-toxic base addition salts, *i.e.*, salts containing pharmacologically acceptable cations such as, but not limited to, alkali metal or alkaline earth metal salts and the calcium, magnesium, sodium or potassium salts in particular. Suitable organic bases include, but are not limited to, N,N-dibenzylethylenediamine, chloroprocaine, choline, diethanolamine, ethylenediamine, meglumaine (N-methylglucamine), lysine, and procaine.

As used herein and unless otherwise indicated, the term "prodrug" means a derivative of a compound that can hydrolyze, oxidize, or otherwise react under biological conditions (*in vitro* or *in vivo*) to provide the compound. Examples of prodrugs include, but are not limited to, derivatives of selective cytokine inhibitory drugs that comprise biohydrolyzable moieties such as biohydrolyzable amides, biohydrolyzable esters, biohydrolyzable carbamates, biohydrolyzable carbonates, biohydrolyzable ureides, and biohydrolyzable phosphate analogues. Other examples of prodrugs include derivatives of a selective cytokine inhibitory drug that comprise -NO, -NO_2, -ONO, or -ONO_2 moieties. Prodrugs can typically be prepared using well-known methods, such as those described in 1 *Burger's Medicinal Chemistry and Drug Discovery*, 172-178, 949-982 (Manfred E. Wolff *ed.*, 5th ed. 1995), and *Design of Prodrugs* (H. Bundgaard ed., Elselvier, New York 1985).

As used herein and unless otherwise indicated, the terms "biohydrolyzable amide," "biohydrolyzable ester," "biohydrolyzable carbamate," "biohydrolyzable carbonate,"

"biohydrolyzable ureide," "biohydrolyzable phosphate" mean an amide, ester, carbamate, carbonate, ureide, or phosphate, respectively, of a compound that either: 1) does not interfere with the biological activity of the compound but can confer upon that compound advantageous properties *in vivo*, such as uptake, duration of action, or onset of action; or 2) is biologically inactive but is converted *in vivo* to the biologically active compound. Examples of biohydrolyzable esters include, but are not limited to, lower alkyl esters, lower acyloxyalkyl esters (such as acetoxylmethyl, acetoxyethyl, aminocarbonyloxymethyl, pivaloyloxymethyl, and pivaloyloxyethyl esters), lactonyl esters (such as phthalidyl and thiophthalidyl esters), lower alkoxyacyloxyalkyl esters (such as methoxycarbonyloxymethyl, ethoxycarbonyloxyethyl and isopropoxycarbonyloxyethyl esters), alkoxyalkyl esters, choline esters, and acylamino alkyl esters (such as acetamidomethyl esters). Examples of biohydrolyzable amides include, but are not limited to, lower alkyl amides, α-amino acid amides, alkoxyacyl amides, and alkylaminoalkylcarbonyl amides. Examples of biohydrolyzable carbamates include, but are not limited to, lower alkylamines, substituted ethylenediamines, aminoacids, hydroxyalkylamines, heterocyclic and heteroaromatic amines, and polyether amines.

Various selective cytokine inhibitory drugs contain one or more chiral centers, and can exist as racemic mixtures of enantiomers or mixtures of diastereomers. This invention encompasses the use of stereomerically pure forms of such compounds, as well as the use of mixtures of those forms. For example, mixtures comprising equal or unequal amounts of the enantiomers of selective cytokine inhibitory drugs may be used in methods and compositions of the invention. The purified (R) or (S) enantiomers of the specific compounds disclosed herein may be used substantially free of its other enantiomer.

As used herein and unless otherwise indicated, the term "stereomerically pure" means a composition that comprises one stereoisomer of a compound and is substantially free of other stereoisomers of that compound. For example, a stereomerically pure composition of a compound having one chiral center will be substantially free of the opposite enantiomer of the compound. A stereomerically pure composition of a compound having two chiral centers will be substantially free of other diastereomers of the compound. A typical stereomerically pure compound comprises greater than about 80% by weight of one stereoisomer of the compound and less than about 20% by weight of other stereoisomers of the compound, more preferably greater than about 90% by weight of one stereoisomer of the compound and less than about 10% by weight of the other stereoisomers of the compound, even more preferably greater than about 95% by weight of one

stereoisomer of the compound and less than about 5% by weight of the other stereoisomers of the compound, and most preferably greater than about 97% by weight of one stereoisomer of the compound and less than about 3% by weight of the other stereoisomers of the compound. As used herein and unless otherwise indicated, the term "stereomerically enriched" means a composition that comprises greater than about 60% by weight of one stereoisomer of a compound, preferably greater than about 70% by weight, more preferably greater than about 80% by weight of one stereoisomer of a compound. As used herein and unless otherwise indicated, the term "enantiomerically pure" means a stereomerically pure composition of a compound having one chiral center. Similarly, the term "stereomerically enriched" means a stereomerically enriched composition of a compound having one chiral center.

It should be noted that if there is a discrepancy between a depicted structure and a name given that structure, the depicted structure is to be accorded more weight. In addition, if the stereochemistry of a structure or a portion of a structure is not indicated with, for example, bold or dashed lines, the structure or portion of the structure is to be interpreted as encompassing all stereoisomers of it.

4.2 SECOND ACTIVE INGREDIENTS

One or more second active ingredients can be used in combination with a selective cytokine inhibitory drug of the present invention. Preferably, the second active ingredient, or agent, is capable of suppressing the overproduction of hematopoietic stem cells, or ameliorating one or more of the symptoms of MPD.

Second active agents can be, but are not limited to, small molecules (*e.g.*, synthetic inorganic, organometallic, or organic molecules), large molecules, synthetic drugs, peptides, polypeptides, proteins, nucleic acids, antibodies and the like. Any agent that is known to be useful, or that has been used or is currently being used for the prevention, treatment or amelioration of one or more symptoms of MPD can be used in the combination with the present invention. Particular agents include, but are not limited to, anticancer agents (*e.g.*, antimetabolites, antibiotics, alkylating agents, microtubule inhibitors, steroid hormones, DNA-repair enzyme inhibitors, kinase inhibitors, farnesyl transferase inhibitors, antisense oligonucleotides, immunomodulators, antibodies, vaccines, and adnosine deaminase inhibitors), all-trans retinoic acid (*e.g.*, arsenic trioxide), platelet inhibitors (*e.g.*, aspirin, dipyridamole, ticlopidine, anagrelide), anticoagulants (*e.g.*, enoxaprin, heparin, warfarin), thrombolytic agents (*e.g.*, alteplase (tPA), anistreplase, streptokinase, urokinase), antifibrosis agents (*e.g.*, penicillamine, suramin, clochicine), agents used in treating

bleeding (*e.g.*, aminocaproic acid, protamine sulfate, vitamin K), and agents used in treating anemia (*e.g.*, vitamin K, folic acid).

This invention also encompasses the use of native, naturally occurring, and recombinant proteins. The invention further encompasses mutants and derivatives (*e.g.*, modified forms) of naturally occurring proteins that exhibit, *in vivo*, at least some of the pharmacological activity of the proteins upon which they are based. Examples of mutants include, but are not limited to, proteins that have one or more amino acid residues that differ from the corresponding residues in the naturally occurring forms of the proteins. Also encompassed by the term "mutants" are proteins that lack carbohydrate moieties normally present in their naturally occurring forms (*e.g.*, nonglycosylated forms). Examples of derivatives include, but are not limited to, pegylated derivatives and fusion proteins, such as proteins formed by fusing IgG1 or IgG3 to the protein or active portion of the protein of interest. *See, e.g.*, Penichet, M.L. and Morrison, S.L., *J. Immunol. Methods* 248:91-101 (2001).

This invention further encompasses the use of immune cells or transplantation of blood and marrow stem cells. For example, CML patients can be treated with infusion of donor white blood cells that suppress the growth of leukemia cells. Slavin *et al.*, *Transfus Apheresis Sci* 27(2):159-66 (2002).

Examples of anti-cancer drugs that can be used in the various embodiments of the invention, including the methods, dosing regimens, cocktails, pharmaceutical compositions and dosage forms and kits of the invention, include, but are not limited to: acivicin; aclarubicin; acodazole hydrochloride; acronine; adozelesin; aldesleukin; altretamine; ambomycin; ametantrone acetate; aminoglutethan immunomodulatory compound of the inventione; amsacrine; anastrozole; anthramycin; asparaginase; asperlin; azacitidine; azetepa; azotomycin; batimastat; benzodepa; bicalutamide; bisantrene hydrochloride; bisnafide dimesylate; bizelesin; bleomycin sulfate; brequinar sodium; bropirimine; busulfan; cactinomycin; calusterone; caracemide; carbetimer; carboplatin; carmustine; carubicin hydrochloride; carzelesin; cedefingol; celecoxib (COX-2 inhibitor); chlorambucil; cirolemycin; cisplatin; cladribine; crisnatol mesylate; cyclophosphamide; cytarabine; dacarbazine; dactinomycin; daunorubicin hydrochloride; decitabine; dexormaplatin; dezaguanine; dezaguanine mesylate; diaziquone; dacarbazine; docetaxel; doxorubicin; doxorubicin hydrochloride; droloxifene; droloxifene citrate; dromostanolone propionate; duazomycin; edatrexate; eflornithine hydrochloride; elsamitrucin; enloplatin; enpromate; epipropidine; epirubicin hydrochloride; erbulozole; esorubicin hydrochloride; estramustine;

estramustine phosphate sodium; etanidazole; etoposide; etoposide phosphate; etoprine; fadrozole hydrochloride; fazarabine; fenretinide; floxuridine; fludarabine phosphate; fluorouracil; flurocitabine; fosquidone; fostriecin sodium; gemcitabine; gemcitabine hydrochloride; hydroxyurea; idarubicin hydrochloride; ifosfamide; ilmofosine; interleukin II (including recombinant interleukin II, or rIL2), interferon alfa-2a; interferon alfa-2b; interferon alfa-n1; interferon alfa-n3; interferon beta-I a; interferon gamma-I b; iproplatin; irinotecan; irinotecan hydrochloride; lanreotide acetate; letrozole; leuprolide acetate; liarozole hydrochloride; lometrexol sodium; lomustine; losoxantrone hydrochloride; masoprocol; maytansine; mechlorethamine hydrochloride; megestrol acetate; melengestrol acetate; melphalan; menogaril; mercaptopurine; methotrexate; methotrexate sodium; metoprine; meturedepa; mitindomide; mitocarcin; mitocromin; mitogillin; mitomalcin; mitomycin; mitosper; mitotane; mitoxantrone hydrochloride; mycophenolic acid; nocodazole; nogalamycin; oblimersen; ormaplatin; oxisuran; paclitaxel; pegaspargase; peliomycin; pentamustine; peplomycin sulfate; perfosfamide; pipobroman; piposulfan; piroxantrone hydrochloride; plicamycin; plomestane; porfimer sodium; porfiromycin; prednimustine; procarbazine hydrochloride; puromycin; puromycin hydrochloride; pyrazofurin; riboprine; rogletan immunomodulatory compound of the inventione; safingol; safingol hydrochloride; semustine; simtrazene; sparfosate sodium; sparsomycin; spirogermanium hydrochloride; spiromustine; spiroplatin; streptonigrin; streptozocin; sulofenur; talisomycin; tecogalan sodium; taxotere; tegafur; teloxantrone hydrochloride; temoporfin; teniposide; teroxirone; testolactone; thiamiprine; thioguanine; thiotepa; tiazofurin; tirapazamine; toremifene citrate; trestolone acetate; triciribine phosphate; trimetrexate; trimetrexate glucuronate; triptorelin; tubulozole hydrochloride; uracil mustard; uredepa; vapreotide; verteporfin; vinblastine sulfate; vincristine sulfate; vindesine; vindesine sulfate; vinepidine sulfate; vinglycinate sulfate; vinleurosine sulfate; vinorelbine tartrate; vinrosidine sulfate; vinzolidine sulfate; vorozole; zeniplatin; zinostatin; zorubicin hydrochloride. Other anti-cancer drugs include, but are not limited to: 20-epi-1,25 dihydroxyvitamin D3; 5-ethynyluracil; abiraterone; aclarubicin; acylfulvene; adecypenol; adozelesin; aldesleukin; ALL-TK antagonists; altretamine; ambamustine; amidox; amifostine; aminolevulinic acid; amrubicin; amsacrine; anagrelide; anastrozole; andrographolide; angiogenesis inhibitors; antagonist D; antagonist G; antarelix; anti-dorsalizing morphogenetic protein-1; antiandrogen, prostatic carcinoma; antiestrogen; antineoplaston; antisense oligonucleotides; aphidicolin glycinate; apoptosis gene modulators; apoptosis regulators; apurinic acid; ara-CDP-DL-PTBA; arginine deaminase;

asulacrine; atamestane; atrimustine; axinastatin 1; axinastatin 2; axinastatin 3; azasetron; azatoxin; azatyrosine; baccatin III derivatives; balanol; batimastat; BCR/ABL antagonists; benzochlorins; benzoylstaurosporine; beta lactam derivatives; beta-alethine; betaclamycin B; betulinic acid; bFGF inhibitor; bicalutamide; bisantrene; bisaziridinylspermine; bisnafide; bistratene A; bizelesin; breflate; bropirimine; budotitane; buthionine sulfoximine; calcipotriol; calphostin C; camptothecin derivatives; canarypox IL-2; capecitabine; carboxamide-amino-triazole; carboxyamidotriazole; CaRest M3; CARN 700; cartilage derived inhibitor; carzelesin; casein kinase inhibitors (ICOS); castanospermine; cecropin B; cetrorelix; chlorlns; chloroquinoxaline sulfonamide; cicaprost; cis-porphyrin; cladribine; clomifene analogues; clotrimazole; collismycin A; collismycin B; combretastatin A4; combretastatin analogue; conagenin; crambescidin 816; crisnatol; cryptophycin 8; cryptophycin A derivatives; curacin A; cyclopentanthraquinones; cycloplatam; cypemycin; cytarabine ocfosfate; cytolytic factor; cytostatin; dacliximab; decitabine; dehydrodidemnin B; deslorelin; dexamethasone; dexifosfamide; dexrazoxane; dexverapamil; diaziquone; didemnin B; didox; diethylnorspermine; dihydro-5-azacytidine; dihydrotaxol, 9-; dioxamycin; diphenyl spiromustine; docetaxel; docosanol; dolasetron; doxifluridine; droloxifene; dronabinol; duocarmycin SA; ebselen; ecomustine; edelfosine; edrecolomab; eflornithine; elemene; emitefur; epirubicin; epristeride; estramustine analogue; estrogen agonists; estrogen antagonists; etanidazole; etoposide phosphate; exemestane; fadrozole; fazarabine; fenretinide; filgrastim; finasteride; flavopiridol; flezelastine; fluasterone; fludarabine; fluorodaunorunicin hydrochloride; forfenimex; formestane; fostriecin; fotemustine; gadolinium texaphyrin; gallium nitrate; galocitabine; ganirelix; gelatinase inhibitors; gemcitabine; glutathione inhibitors; hepsulfam; heregulin; hexamethylene bisacetamide; hypericin; ibandronic acid; idarubicin; idoxifene; idramantone; ilmofosine; ilomastat; an immunomodulatory compound of the inventionazoacridones; imiquimod; immunostimulant peptides; insulin-like growth factor-1 receptor inhibitor; interferon agonists; interferons; interleukins; iobenguane; iododoxorubicin; ipomeanol, 4-; iroplact; irsogladine; isobengazole; isohomohalicondrin B; itasetron; jasplakinolide; kahalalide F; lamellarin-N triacetate; lanreotide; leinamycin; lenograstim; lentinan sulfate; leptolstatin; letrozole; leukemia inhibiting factor; leukocyte alpha interferon; leuprolide+estrogen+progesterone; leuprorelin; levamisole; liarozole; linear polyamine analogue; lipophilic disaccharide peptide; lipophilic platinum compounds; lissoclinamide 7; lobaplatin; lombricine; lometrexol; lonidamine; losoxantrone; lovastatin; loxoribine; lurtotecan; lutetium texaphyrin; lysofylline; lytic peptides; maitansine; mannostatin A;

marimastat; masoprocol; maspin; matrilysin inhibitors; matrix metalloproteinase inhibitors; menogaril; merbarone; meterelin; methioninase; metoclopramide; MIF inhibitor; mifepristone; miltefosine; mirimostim; mismatched double stranded RNA; mitoguazone; mitolactol; mitomycin analogues; mitonafide; mitotoxin fibroblast growth factor-saporin; mitoxantrone; mofarotene; molgramostim; monoclonal antibody, human chorionic gonadotrophin; monophosphoryl lipid A+myobacterium cell wall sk; mopidamol; multiple drug resistance gene inhibitor; multiple tumor suppressor 1-based therapy; mustard anticancer agent; mycaperoxide B; mycobacterial cell wall extract; myriaporone; N-acetyldinaline; N-substituted benzamides; nafarelin; nagrestip; naloxone+pentazocine; napavin; naphterpin; nartograstim; nedaplatin; nemorubicin; neridronic acid; neutral endopeptidase; nilutamide; nisamycin; nitric oxide modulators; nitroxide antioxidant; nitrullyn; O^6-benzylguanine; octreotide; okicenone; oligonucleotides; onapristone; ondansetron; ondansetron; oracin; oral cytokine inducer; ormaplatin; osaterone; oxaliplatin; oxaunomycin; paclitaxel; paclitaxel analogues; paclitaxel derivatives; palauamine; palmitoylrhizoxin; pamidronic acid; panaxytriol; panomifene; parabactin; pazelliptine; pegaspargase; peldesine; pentosan polysulfate sodium; pentostatin; pentrozole; perflubron; perfosfamide; perillyl alcohol; phenazinomycin; phenylacetate; phosphatase inhibitors; picibanil; pilocarpine hydrochloride; pirarubicin; piritrexim; placetin A; placetin B; plasminogen activator inhibitor; platinum complex; platinum compounds; platinum-triamine complex; porfimer sodium; porfiromycin; prednisone; propyl bis-acridone; prostaglandin J2; proteasome inhibitors; protein A-based immune modulator; protein kinase C inhibitor; protein kinase C inhibitors, microalgal; protein tyrosine phosphatase inhibitors; purine nucleoside phosphorylase inhibitors; purpurins; pyrazoloacridine; pyridoxylated hemoglobin polyoxyethylene conjugate; raf antagonists; raltitrexed; ramosetron; ras farnesyl protein transferase inhibitors; ras inhibitors; ras-GAP inhibitor; retelliptine demethylated; rhenium Re 186 etidronate; rhizoxin; ribozymes; RII retinamide; rogletan immunomodulatory compound of the inventione; rohitukine; romurtide; roquinimex; rubiginone B1; ruboxyl; safingol; saintopin; SarCNU; sarcophytol A; sargramostim; Sdi 1 mimetics; semustine; senescence derived inhibitor 1; sense oligonucleotides; signal transduction inhibitors; signal transduction modulators; single chain antigen binding protein; sizofiran; sobuzoxane; sodium borocaptate; sodium phenylacetate; solverol; somatomedin binding protein; sonermin; sparfosic acid; spicamycin D; spiromustine; splenopentin; spongistatin 1; squalamine; stem cell inhibitor; stem-cell division inhibitors; stipiamide; stromelysin inhibitors; sulfinosine; superactive vasoactive

intestinal peptide antagonist; suradista; suramin; swainsonine; synthetic glycosaminoglycans; tallimustine; tamoxifen methiodide; tauromustine; tazarotene; tecogalan sodium; tegafur; tellurapyrylium; telomerase inhibitors; temoporfin; teniposide; tetrachlorodecaoxide; tetrazomine; thaliblastine; thiocoraline; thrombopoietin; thrombopoietin mimetic; thymalfasin; thymopoietin receptor agonist; thymotrinan; thyroid stimulating hormone; tin ethyl etiopurpurin; tirapazamine; titanocene bichloride; topsentin; toremifene; totipotent stem cell factor; translation inhibitors; tretinoin; triacetyluridine; triciribine; trimetrexate; triptorelin; tropisetron; turosteride; tyrosine kinase inhibitors; tyrphostins; UBC inhibitors; ubenimex; urogenital sinus-derived growth inhibitory factor; urokinase receptor antagonists; vapreotide; variolin B; vector system, erythrocyte gene therapy; velaresol; veramine; verdins; verteporfin; vinorelbine; vinxaltine; vitaxin; vorozole; zanoterone; zeniplatin; zilascorb; and zinostatin stimalamer. Preferred anti-cancer drugs are those that have been shown to have treatment benefit in a MPD patient, *e.g.*, interferon-α, hydroxyurea, busulfan, anagrelide, daunorubicin, cincristine, corticosteroid hormones (*e.g.*, prednisone, beclomethasone, cortisone, dexamethasone, fludrocortisone, hydrocortisone, methylprednisolone), kinase inhibitors, topoisomerase inhibitors, farnesyl transferase inhibitors, vaccines and antisense nucleotides.

Examples of kinase inhibitors include, but are not limited to, compound ST1571, imatinib mesylate (Kantarjian *et al.*, *Clin Cancer Res.* 8(7):2167-76 (2002)), and those compounds disclosed in U.S. Pat. Nos. 6,245,759, 6,399,633, 6,383,790, 6,335,156, 6,271,242, 6,242,196, 6,218,410, 6,218,372, 6,057,300, 6,034,053, 5,985,877, 5,958,769, 5,925,376, 5,922,844, 5,911,995, 5,872,223, 5,863,904, 5,840,745, 5,728,868, 5,648,239, 5,587,459, all of which are incorporated herein by reference. Preferred kinase inhibitors include, but are not limited to, those that directly target the BCR/ABL kinase or other kinases that are involved in the MPD pathophysiology, *e.g.*, ST1571, and imatinib mesylate.

Examples of topoisomerase inhibitors include, but are not limited to, camptothecin; irinotecan; SN-38; topotecan; 9-aminocamptothecin; GG-211 (GI 147211); DX-8951f; IST-622; rubitecan; pyrazoloacridine; XR-5000; saintopin; UCE6; UCE1022; TAN-1518A; TAN-1518B; KT6006; KT6528; ED-110; NB-506; ED-110; NB-506; and rebeccamycin; bulgarein; DNA minor groove binders such as Hoescht dye 33342 and Hoechst dye 33258; nitidine; fagaronine; epiberberine; coralyne; beta-lapachone; BC-4-1; and pharmaceutically acceptable salts, solvates, clathrates, and prodrugs thereof. *See, e.g.*, Rothenberg, M.L., *Annals of Oncology* 8:837-855(1997); and Moreau, P., *et al.*, *J. Med. Chem.* 41:1631-1640(1998). Examples of camptothecin derivatives that can be used in the methods and

compositions of this invention are disclosed by, for example, U.S. Patent Nos.: 6,043,367; 6,040,313; 5,932,588; 5,916,896; 5,889,017; 5,801,167; 5,674,874; 5,658,920; 5,646,159; 5,633,260; 5,604,233; 5,597,829; 5,552,154; 5,541,327; 5,525,731; 5,468,754; 5,447,936; 5,446,047; 5,401,747; 5,391,745; 5,364,858; 5,340,817; 5,244,903; 5,227,380; 5,225,404; 5,180,722; 5,122,606; 5,122,526; 5,106,742; 5,061,800; 5,053,512; 5,049,668; 5,004,758; 4,981,968; 4,943,579; 4,939,255; 4,894,456; and 4,604,463, each of which is incorporated herein by reference. Preferred topoisomerase inhibitors include, but are not limited to, DX-8951f, irinotecan, SN-38, and pharmaceutically acceptable salts, solvates, clathrates, and prodrugs thereof.

Examples of farnesyl transferase inhibitor include, but are not limited to, R115777, BMS-214662, (for review, see Caponigro, *Anticancer Drugs* 13(8):891-897 (2002)), and those disclosed by, for example, U.S. Patent Nos: 6,458,935, 6,451,812, 6,440,974, 6,436,960, 6,432,959, 6,420,387, 6,414,145, 6,410,541, 6,410,539, 6,403,581, 6,399,615, 6,387,905, 6,372,747, 6,369,034, 6,362,188, 6,342,765, 6,342,487, 6,300,501, 6,268,363, 6,265,422, 6,248,756, 6,239,140, 6,232,338, 6,228,865, 6,228,856, 6,225,322, 6,218,406, 6,211,193, 6,187,786, 6,169,096, 6,159,984, 6,143,766, 6,133,303, 6,127,366, 6,124,465, 6,124,295, 6,103,723, 6,093,737, 6,090,948, 6,080,870, 6,077,853, 6,071,935, 6,066,738, 6,063,930, 6,054,466, 6,051,582, 6,051,574, 6,040,305, all of which are incorporated herein by reference.

In one embodiment of the present invention, the second active agent is an agent used in the gene therapy of MPD. For example, antisense oligonucleotides can block the encoding instructions of an oncogene so that it cannot direct the formation of the corresponding oncoprotein that causes the cell to transform into a malignant cell. Examples of antisense oligonucleotides include, but are not limited to, those disclosed in the U.S. Pat. Nos. 6,277,832, 5,998,596, 5,885,834, 5,734,033, and 5,618,709, all of which are incorporated herein by reference.

In another embodiment of the present invention, the second active agent is a protein, a fusion protein thereof, or a vaccine that secretes the protein, wherein the protein is IL-2, IL-10, IL-12, IL18, G-CSF, GM-CSF, EPO, or a pharmacologically active mutant or derivative thereof. In some circumstances apparent to one skilled in the art, G-CSF, GM-CSF and EPO are not preferred. For example, G-CSF, GM-CSF and EPO preferably are not used in methods that do not utilize stem cell transplantation. In a preferred embodiment, the protein is an antibody or an antibody linked to a chemical toxin or radioactive isotope that targets and kills specific overproduced cells in a MPD patient.

Such antibodies include, but are not limited to, rituximab (Rituxan®), calicheamycin (Mylotarg®), ibritumomab tiuxetan (Zevalin®), and tositumomab (Bexxar®).

In a specific embodiment of the present invention, the second active agent is a vaccine that can induce antigen-specific anti-malignant cell immune responses in a MPD patient. A non-limiting example of such a vaccine is disclosed in U.S. Pat. No. 6,432,925, which is incorporated herein by reference.

In yet another embodiment of the present invention, the second active agent is one that is capable of reversal of multidrug resistance in MPD patients. The overproduced cells in MPD patients have mechanisms that may allow them to escape the damaging effects of chemotherapy. New agents are being studied to decrease resistance to an important chemotherapeutic drug used in the treatment of leukemia. Non-limiting examples of such agents are disclosed in U.S. Pat. No. 6,225,325, which is incorporated herein by reference.

Other agents that can be used in combination with the present invention include, but are not limited to those disclosed in U.S. Pat Nos. 6,096,300, 6,420,391,6,326,205, 5,866,332, 6,458,349, 6,420,378, 6,399,664, 6,395,771, 6,346,246, 6,333,309, 6,331,642, 6,329,497, 6,326,378, 6,313,129, 6,306,393, 6,303,646, 6,265,427, 6,262,053, 6,258,779, 6,251,882, 6,231,893, 6,225,323, 6,221,873, 6,218,412, 6,204,364, 6,187,287, 6,183,988, 6,183,744, 6,172,112, 6,156,733, 6,143,738, 6,127,406, 6,121,320, 6,107,520, 6,107,457, 6,075,015, and 6,063,814, all of which are incorporated herein by reference.

4.3 METHODS OF TREATMENT AND MANAGEMENT

Methods of this invention encompass methods of preventing, treating and/or managing various types of MPD. As used herein, unless otherwise specified, the terms "treating" and "preventing" encompass the inhibition or the reduction of the severity or magnitude of one or more symptoms or laboratory findings associated with MPD. Symptoms associated with MPD include, but are not limited to, headache, dizziness, tinnitus, blurred vision, fatigue, night sweat, low-grade fever, generalized pruritus, epistaxis, blurred vision, splenomegaly, abdominal fullness, thrombosis, increased bleeding, anemia, splenic infarction, severe bone pain, hematopoiesis in the liver, ascites, esophageal varices, liver failure, respiratory distress, and priapism. Laboratory findings associated with MPD include, but are not limited to, clonal expansion of a multipotent hematopoietic progenitor cell with the overproduction of one or more of the formed elements of the blood (*e.g.*, elevated red blood cell count, elevated white blood cell count, and/or elevated platelet count), presence of Philadelphia chromosome or bcr-abl gene, teardrop poikilocytosis on peripheral blood smear, leukoerythroblastic blood pictuer, giant abnormal platelets,

hypercellular bone marrow with reticular or collagen fibrosis, and marked left-shifted myeloid series with a low percentage of promyelocytes and blasts. As used herein, unless otherwise specified, the term "treating" refers to the administration of a composition after the onset of symptoms of MPD, whereas "preventing" refers to the administration prior to the onset of symptoms, particularly to patients at risk of MPD. As used herein and unless otherwise indicated, the term "managing" encompasses preventing the recurrence of MPD in a patient who had suffered from MPD, lengthening the time a patient who had suffered from MPD remains in remission, and/or preventing the occurrence of MPD in patients at risk of suffering from MPD.

The invention encompasses methods of treating or preventing patients with primary and secondary MPD. It further encompasses methods treating patients who have been previously treated for MPD, as well as those who have not previously been treated for MPD. Because patients with MPD have heterogenous clinical manifestations and varying clinical outcomes, it has become apparent that staging the patients according to their prognosis and approaching therapy depending on the severity and stage may be necessary. Indeed, the methods and compositions of this invention can be used in various stages of treatments for patients with one or more types of MPD including, but not limited to, polycythemia rubra vera (PRV), primary thromobocythemia (PT), chronic myelogenous leukemia (CML), and agnogenic myeloid metaplasia (AMM).

Methods encompassed by this invention comprise administering a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof to a patient (*e.g.*, a human) suffering, or likely to suffer, from MPD. Specific patient populations include the elderly, *i.e.*, ages 60 and above as well as those over 35 years of age. Patients with familial history of MPD or leukemia are also preferred candidates for preventive regimens.

In one embodiment of the invention, the recommended daily dose range of a selective cytokine inhibitory drug for the conditions described herein lie within the range of from about 1 mg to about 10,000 mg per day, given as a single once-a-day dose, or preferably in divided doses throughout a day. More specifically, the daily dose is administered twice daily in equally divided doses. Specifically, a daily dose range should be from about 1 mg to about 5,000 mg per day, more specifically, between about 10 mg and about 2,500 mg per day, between about 100 mg and about 800 mg per day, between about100 mg and about1,200 mg per day, or between about 25 mg and about 2,500 mg per day. In managing the patient, the therapy should be initiated at a lower dose, perhaps about

1 mg to about 2,500 mg, and increased if necessary up to about 200 mg to about 5,000 mg per day as either a single dose or divided doses, depending on the patient's global response. In a particular embodiment, 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide can be preferably administered in an amount of about 400, 800, 1,200, 2,500, 5,000 or 10,000 mg a day as two divided doses.

4.3.1 Combination Therapy With A Second Active Agent

Particular methods of the invention comprise administering 1) a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, and 2) a second active agent or active ingredient. Examples of selective cytokine inhibitory drugs of the invention are disclosed herein (*see, e.g.*, section 4.1); and examples of the second active agents are also disclosed herein (*see, e.g.*, section 4.2).

In particular embodiments, one or more selective cytokine inhibitory drugs are administered in combination with the administration of one or more therapies that are used to treat, manage, or prevent myeloproliferative diseases. A non-limiting example is the use of selective cytokine inhibitory drugs of the invention in combination with the administration of an anti-cancer cocktail regimen, such as, but not limited to, a regimen that includes cytarabine and an anthracycline (*e.g.*, daunorubicin or idarubicin).

Administration of the selective cytokine inhibitory drugs and the second active agents to a patient can occur simultaneously or sequentially by the same or different routes of administration. The suitability of a particular route of administration employed for a particular active agent will depend on the active agent itself (*e.g.*, whether it can be administered orally without decomposing prior to entering the blood stream) and the disease being treated. A preferred route of administration for a selective cytokine inhibitory drug is oral. Preferred routes of administration for the second active agents or ingredients of the invention are known to those of ordinary skill in the art. *See, e.g.*, *Physicians' Desk Reference*, 1755-1760 (56^{th} ed., 2002).

In one embodiment, the second active agent is administered intravenously or subcutaneously and once or twice daily in an amount of from about 1 to about 1000 mg, from about 5 to about 500 mg, from about 10 to about 350 mg, or from about 50 to about 200 mg. The specific amount of the second active agent will depend on the specific agent used, the type of MPD being treated or managed, the severity and stage of MPD, and the amount(s) of selective cytokine inhibitory drugs of the invention and any optional additional active agents concurrently administered to the patient. In a particular

embodiment, the second active agent is interferon-α, hydroxyurea, anagrelide, arsenic troxide, ST1571, imatinib mesylate, DX-8951f, R115777, vincristine, daunorubicin, prednisone or a combination thereof. Interferon-α is administered in an amount of from 2 to 5 million unites subcutaneously three times weekly. Hydroxyurea is administered in an amount of from about 500 to about 1500 mg/d orally, adjusted to keep platelets less than 500,000/μL without reducing the neutrophil count to < 2000/μL.

4.3.2 Use With Transplantation Therapy

In still another embodiment, this invention encompasses a method of treating, preventing and/or managing MPD, which comprises administering the selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, in conjunction with transplantation therapy. As discussed elsewhere herein, the treatment of MPD is based on the stages and mechanism of the disease. As inevitable leukemic transformation develops in certain stages of MPD, transplantation of peripheral blood stem cells, hematopoietic stem cell preparation or bone marrow may be necessary. The combined use of the selective cytokine inhibitory drug of the invention and transplantation therapy provides a unique and unexpected synergism. In particular, a selective cytokine inhibitory drug of the invention exhibits immunomodulatory activity that may provide additive or synergistic effects when given concurrently with transplantation therapy in patients with MPD. A selective cytokine inhibitory drug of the invention can work in combination with transplantation therapy reducing complications associated with the invasive procedure of transplantation and risk of related Graft Versus Host Disease (GVHD). This invention encompasses a method of treating, preventing and/or managing MPD which comprises administering to a patient (*e.g.*, a human) a selective cytokine inhibitory drug of the invention, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, before, during, or after the transplantation of umbilical cord blood, placental blood, peripheral blood stem cell, hematopoietic stem cell preparation or bone marrow. Examples of stem cells suitable for use in the methods of the invention are disclosed in U.S. provisional patent application no. 60/372,348, filed April 12, 2002 by R. Hariri *et al.*, the entirety of which is incorporated herein by reference.

4.3.3 Cycling Therapy

In certain embodiments, the prophylactic or therapeutic agents of the invention are cyclically administered to a patient. Cycling therapy involves the administration of an

active agent for a period of time, followed by a rest for a period of time, and repeating this sequential administration. Cycling therapy can reduce the development of resistance to one or more of the therapies, avoid or reduce the side effects of one of the therapies, and/or improves the efficacy of the treatment.

Consequently, in one specific embodiment of the invention, a selective cytokine inhibitory drug of the invention is administered daily in a single or divided doses in a four to six week cycle with a rest period of about a week or two weeks. The invention further allows the frequency, number, and length of dosing cycles to be increased. Thus, another specific embodiment of the invention encompasses the administration of a selective cytokine inhibitory drug of the invention for more cycles than are typical when it is administered alone. In yet another specific embodiment of the invention, a selective cytokine inhibitory drug of the invention is administered for a greater number of cycles that would typically cause dose-limiting toxicity in a patient to whom a second active ingredient is not also being administered.

In one embodiment, a selective cytokine inhibitory drug of the invention is administered daily and continuously for three or four weeks at a dose of from about 0.1 to about 150 mg/d followed by a break of one or two weeks.

In one embodiment of the invention a selective cytokine inhibitory drug of the invention and a second active ingredient are administered orally, with administration of a selective cytokine inhibitory drug of the invention occurring 30 to 60 minutes prior to a second active ingredient, during a cycle of four to six weeks. In another embodiment of the invention, the combination of a selective cytokine inhibitory drug of the invention and a second active ingredient is administered by intravenous infusion over about 90 minutes every cycle. Typically, the number of cycles during which the combinatorial treatment is administered to a patient will be from about one to about 24 cycles, more typically from about two to about 16 cycles, and even more typically from about four to about eight cycles.

4.4 PHARMACEUTICAL COMPOSITIONS AND SINGLE UNIT DOSAGE FORMS

Pharmaceutical compositions can be used in the preparation of individual, single unit dosage forms. Pharmaceutical compositions and dosage forms of the invention comprise a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof. Pharmaceutical compositions and dosage forms of the invention can further comprise one or more excipients.

Pharmaceutical compositions and dosage forms of the invention can also comprise one or more additional active ingredients. Consequently, pharmaceutical compositions and dosage forms of the invention comprise the active ingredients disclosed herein (*e.g.*, a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof, and a second active ingredient). Examples of optional additional active ingredients are disclosed herein (*see, e.g.*, section 4.2).

Single unit dosage forms of the invention are suitable for oral, mucosal (*e.g.*, nasal, sublingual, vaginal, buccal, or rectal), or parenteral (*e.g.*, subcutaneous, intravenous, bolus injection, intramuscular, or intraarterial), transdermal or transcutaneous administration to a patent. Examples of dosage forms include, but are not limited to: tablets; caplets; capsules, such as soft elastic gelatin capsules; cachets; troches; lozenges; dispersions; suppositories; powders; aerosols (*e.g.*, nasal sprays or inhalers); gels; liquid dosage forms suitable for oral or mucosal administration to a patient, including suspensions (*e.g.*, aqueous or non-aqueous liquid suspensions, oil-in-water emulsions, or a water-in-oil liquid emulsions), solutions, and elixirs; liquid dosage forms suitable for parenteral administration to a patient; and sterile solids (*e.g.*, crystalline or amorphous solids) that can be reconstituted to provide liquid dosage forms suitable for parenteral administration to a patient.

The composition, shape, and type of dosage forms of the invention will typically vary depending on their use. For example, a dosage form used in the acute treatment of a disease may contain larger amounts of one or more of the active ingredients it comprises than a dosage form used in the chronic treatment of the same disease. Similarly, a parenteral dosage form may contain smaller amounts of one or more of the active ingredients it comprises than an oral dosage form used to treat the same disease. These and other ways in which specific dosage forms encompassed by this invention will vary from one another will be readily apparent to those skilled in the art. *See, e.g., Remington's Pharmaceutical Sciences*, 18th ed., Mack Publishing, Easton PA (1990).

Typical pharmaceutical compositions and dosage forms comprise one or more excipients. Suitable excipients are well known to those skilled in the art of pharmacy, and non-limiting examples of suitable excipients are provided herein. Whether a particular excipient is suitable for incorporation into a pharmaceutical composition or dosage form depends on a variety of factors well known in the art including, but not limited to, the way in which the dosage form will be administered to a patient. For example, oral dosage forms such as tablets may contain excipients not suited for use in parenteral dosage forms. The suitability of a particular excipient may also depend on the specific active ingredients in the

dosage form. For example, the decomposition of some active ingredients may be accelerated by some excipients such as lactose, or when exposed to water. Active ingredients that comprise primary or secondary amines are particularly susceptible to such accelerated decomposition. Consequently, this invention encompasses pharmaceutical compositions and dosage forms that contain little, if any, lactose other mono- or di-saccharides. As used herein, the term "lactose-free" means that the amount of lactose present, if any, is insufficient to substantially increase the degradation rate of an active ingredient.

Lactose-free compositions of the invention can comprise excipients that are well known in the art and are listed, for example, in the *U.S. Pharmacopeia* (USP) 25-NF20 (2002). In general, lactose-free compositions comprise active ingredients, a binder/filler, and a lubricant in pharmaceutically compatible and pharmaceutically acceptable amounts. Preferred lactose-free dosage forms comprise active ingredients, microcrystalline cellulose, pre-gelatinized starch, and magnesium stearate.

This invention further encompasses anhydrous pharmaceutical compositions and dosage forms comprising active ingredients, since water can facilitate the degradation of some compounds. For example, the addition of water (*e.g.*, 5%) is widely accepted in the pharmaceutical arts as a means of simulating long-term storage in order to determine characteristics such as shelf-life or the stability of formulations over time. *See, e.g.*, Jens T. Carstensen, *Drug Stability: Principles & Practice*, 2d. Ed., Marcel Dekker, NY, NY, 1995, pp. 379-80. In effect, water and heat accelerate the decomposition of some compounds. Thus, the effect of water on a formulation can be of great significance since moisture and/or humidity are commonly encountered during manufacture, handling, packaging, storage, shipment, and use of formulations.

Anhydrous pharmaceutical compositions and dosage forms of the invention can be prepared using anhydrous or low moisture containing ingredients and low moisture or low humidity conditions. Pharmaceutical compositions and dosage forms that comprise lactose and at least one active ingredient that comprises a primary or secondary amine are preferably anhydrous if substantial contact with moisture and/or humidity during manufacturing, packaging, and/or storage is expected.

An anhydrous pharmaceutical composition should be prepared and stored such that its anhydrous nature is maintained. Accordingly, anhydrous compositions are preferably packaged using materials known to prevent exposure to water such that they can be included in suitable formulary kits. Examples of suitable packaging include, but are not

limited to, hermetically sealed foils, plastics, unit dose containers (*e.g.*, vials), blister packs, and strip packs.

The invention further encompasses pharmaceutical compositions and dosage forms that comprise one or more compounds that reduce the rate by which an active ingredient will decompose. Such compounds, which are referred to herein as "stabilizers," include, but are not limited to, antioxidants such as ascorbic acid, pH buffers, or salt buffers.

Like the amounts and types of excipients, the amounts and specific types of active ingredients in a dosage form may differ depending on factors such as, but not limited to, the route by which it is to be administered to patients. However, typical dosage forms of the invention comprise a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof in an amount of from about 1 to about 1,200 mg. Typical dosage forms comprise a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, clathrate, or prodrug thereof in an amount of about 1, 2, 5, 10, 25, 50, 100, 200, 400, 800, 1,200, 2,500, 5,000 or 10,000 mg. In a particular embodiment, a preferred dosage form comprises 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide in an amount of about 400, 800 or 1,200 mg. Typical dosage forms comprise the second active ingredient in an amount of 1 to about 1000 mg, from about 5 to about 500 mg, from about 10 to about 350 mg, or from about 50 to about 200 mg. Of course, the specific amount of the second active ingredient will depend on the specific agent used, the type of MPD being treated or managed, and the amount(s) of selective cytokine inhibitory drugs and any optional additional active agents concurrently administered to the patient.

4.4.1 <u>ORAL DOSAGE FORMS</u>

Pharmaceutical compositions of the invention that are suitable for oral administration can be presented as discrete dosage forms, such as, but are not limited to, tablets (*e.g.*, chewable tablets), caplets, capsules, and liquids (*e.g.*, flavored syrups). Such dosage forms contain predetermined amounts of active ingredients, and may be prepared by methods of pharmacy well known to those skilled in the art. *See generally, Remington's Pharmaceutical Sciences*, 18th ed., Mack Publishing, Easton PA (1990).

Typical oral dosage forms of the invention are prepared by combining the active ingredients in an intimate admixture with at least one excipient according to conventional pharmaceutical compounding techniques. Excipients can take a wide variety of forms depending on the form of preparation desired for administration. For example, excipients suitable for use in oral liquid or aerosol dosage forms include, but are not limited to, water,

glycols, oils, alcohols, flavoring agents, preservatives, and coloring agents. Examples of excipients suitable for use in solid oral dosage forms (*e.g.*, powders, tablets, capsules, and caplets) include, but are not limited to, starches, sugars, micro-crystalline cellulose, diluents, granulating agents, lubricants, binders, and disintegrating agents.

Because of their ease of administration, tablets and capsules represent the most advantageous oral dosage unit forms, in which case solid excipients are employed. If desired, tablets can be coated by standard aqueous or nonaqueous techniques. Such dosage forms can be prepared by any of the methods of pharmacy. In general, pharmaceutical compositions and dosage forms are prepared by uniformly and intimately admixing the active ingredients with liquid carriers, finely divided solid carriers, or both, and then shaping the product into the desired presentation if necessary.

For example, a tablet can be prepared by compression or molding. Compressed tablets can be prepared by compressing in a suitable machine the active ingredients in a free-flowing form such as powder or granules, optionally mixed with an excipient. Molded tablets can be made by molding in a suitable machine a mixture of the powdered compound moistened with an inert liquid diluent.

Examples of excipients that can be used in oral dosage forms of the invention include, but are not limited to, binders, fillers, disintegrants, and lubricants. Binders suitable for use in pharmaceutical compositions and dosage forms include, but are not limited to, corn starch, potato starch, or other starches, gelatin, natural and synthetic gums such as acacia, sodium alginate, alginic acid, other alginates, powdered tragacanth, guar gum, cellulose and its derivatives (*e.g.*, ethyl cellulose, cellulose acetate, carboxymethyl cellulose calcium, sodium carboxymethyl cellulose), polyvinyl pyrrolidone, methyl cellulose, pre-gelatinized starch, hydroxypropyl methyl cellulose, (*e.g.*, Nos. 2208, 2906, 2910), microcrystalline cellulose, and mixtures thereof.

Suitable forms of microcrystalline cellulose include, but are not limited to, the materials sold as AVICEL-PH-101, AVICEL-PH-103 AVICEL RC-581, AVICEL-PH-105 (available from FMC Corporation, American Viscose Division, Avicel Sales, Marcus Hook, PA), and mixtures thereof. An specific binder is a mixture of microcrystalline cellulose and sodium carboxymethyl cellulose sold as AVICEL RC-581. Suitable anhydrous or low moisture excipients or additives include AVICEL-PH-103™ and Starch 1500 LM.

Examples of fillers suitable for use in the pharmaceutical compositions and dosage forms disclosed herein include, but are not limited to, talc, calcium carbonate (*e.g.*, granules or powder), microcrystalline cellulose, powdered cellulose, dextrates, kaolin, mannitol,

silicic acid, sorbitol, starch, pre-gelatinized starch, and mixtures thereof. The binder or filler in pharmaceutical compositions of the invention is typically present in from about 50 to about 99 weight percent of the pharmaceutical composition or dosage form.

Disintegrants are used in the compositions of the invention to provide tablets that disintegrate when exposed to an aqueous environment. Tablets that contain too much disintegrant may disintegrate in storage, while those that contain too little may not disintegrate at a desired rate or under the desired conditions. Thus, a sufficient amount of disintegrant that is neither too much nor too little to detrimentally alter the release of the active ingredients should be used to form solid oral dosage forms of the invention. The amount of disintegrant used varies based upon the type of formulation, and is readily discernible to those of ordinary skill in the art. Typical pharmaceutical compositions comprise from about 0.5 to about 15 weight percent of disintegrant, preferably from about 1 to about 5 weight percent of disintegrant.

Disintegrants that can be used in pharmaceutical compositions and dosage forms of the invention include, but are not limited to, agar-agar, alginic acid, calcium carbonate, microcrystalline cellulose, croscarmellose sodium, crospovidone, polacrilin potassium, sodium starch glycolate, potato or tapioca starch, other starches, pre-gelatinized starch, other starches, clays, other algins, other celluloses, gums, and mixtures thereof.

Lubricants that can be used in pharmaceutical compositions and dosage forms of the invention include, but are not limited to, calcium stearate, magnesium stearate, mineral oil, light mineral oil, glycerin, sorbitol, mannitol, polyethylene glycol, other glycols, stearic acid, sodium lauryl sulfate, talc, hydrogenated vegetable oil (*e.g.*, peanut oil, cottonseed oil, sunflower oil, sesame oil, olive oil, corn oil, and soybean oil), zinc stearate, ethyl oleate, ethyl laureate, agar, and mixtures thereof. Additional lubricants include, for example, a syloid silica gel (AEROSIL200, manufactured by W.R. Grace Co. of Baltimore, MD), a coagulated aerosol of synthetic silica (marketed by Degussa Co. of Plano, TX), CAB-O-SIL (a pyrogenic silicon dioxide product sold by Cabot Co. of Boston, MA), and mixtures thereof. If used at all, lubricants are typically used in an amount of less than about 1 weight percent of the pharmaceutical compositions or dosage forms into which they are incorporated.

A preferred solid oral dosage form of the invention comprises a selective cytokine inhibitory drug, anhydrous lactose, microcrystalline cellulose, polyvinylpyrrolidone, stearic acid, colloidal anhydrous silica, and gelatin.

4.4.2 DELAYED RELEASE DOSAGE FORMS

Active ingredients of the invention can be administered by controlled release means or by delivery devices that are well known to those of ordinary skill in the art. Examples include, but are not limited to, those described in U.S. Patent Nos.: 3,845,770; 3,916,899; 3,536,809; 3,598,123; and 4,008,719, 5,674,533, 5,059,595, 5,591,767, 5,120,548, 5,073,543, 5,639,476, 5,354,556, and 5,733,566, each of which is incorporated herein by reference. Such dosage forms can be used to provide slow or controlled-release of one or more active ingredients using, for example, hydropropylmethyl cellulose, other polymer matrices, gels, permeable membranes, osmotic systems, multilayer coatings, microparticles, liposomes, microspheres, or a combination thereof to provide the desired release profile in varying proportions. Suitable controlled-release formulations known to those of ordinary skill in the art, including those described herein, can be readily selected for use with the active ingredients of the invention. The invention thus encompasses single unit dosage forms suitable for oral administration such as, but not limited to, tablets, capsules, gelcaps, and caplets that are adapted for controlled-release.

All controlled-release pharmaceutical products have a common goal of improving drug therapy over that achieved by their non-controlled counterparts. Ideally, the use of an optimally designed controlled-release preparation in medical treatment is characterized by a minimum of drug substance being employed to cure or control the condition in a minimum amount of time. Advantages of controlled-release formulations include extended activity of the drug, reduced dosage frequency, and increased patient compliance. In addition, controlled-release formulations can be used to affect the time of onset of action or other characteristics, such as blood levels of the drug, and can thus affect the occurrence of side (*e.g.*, adverse) effects.

Most controlled-release formulations are designed to initially release an amount of drug (active ingredient) that promptly produces the desired therapeutic effect, and gradually and continually release of other amounts of drug to maintain this level of therapeutic or prophylactic effect over an extended period of time. In order to maintain this constant level of drug in the body, the drug must be released from the dosage form at a rate that will replace the amount of drug being metabolized and excreted from the body. Controlled-release of an active ingredient can be stimulated by various conditions including, but not limited to, pH, temperature, enzymes, water, or other physiological conditions or compounds.

4.4.3 PARENTERAL DOSAGE FORMS

Parenteral dosage forms can be administered to patients by various routes including, but not limited to, subcutaneous, intravenous (including bolus injection), intramuscular, and intraarterial. Because their administration typically bypasses patients' natural defenses against contaminants, parenteral dosage forms are preferably sterile or capable of being sterilized prior to administration to a patient. Examples of parenteral dosage forms include, but are not limited to, solutions ready for injection, dry products ready to be dissolved or suspended in a pharmaceutically acceptable vehicle for injection, suspensions ready for injection, and emulsions.

Suitable vehicles that can be used to provide parenteral dosage forms of the invention are well known to those skilled in the art. Examples include, but are not limited to: Water for Injection USP; aqueous vehicles such as, but not limited to, Sodium Chloride Injection, Ringer's Injection, Dextrose Injection, Dextrose and Sodium Chloride Injection, and Lactated Ringer's Injection; water-miscible vehicles such as, but not limited to, ethyl alcohol, polyethylene glycol, and polypropylene glycol; and non-aqueous vehicles such as, but not limited to, corn oil, cottonseed oil, peanut oil, sesame oil, ethyl oleate, isopropyl myristate, and benzyl benzoate.

Compounds that increase the solubility of one or more of the active ingredients disclosed herein can also be incorporated into the parenteral dosage forms of the invention. For example, cyclodextrin and its derivatives can be used to increase the solubility of a selective cytokine inhibitory drug and its derivatives. *See, e.g.*, U.S. Patent No. 5,134,127, which is incorporated herein by reference.

4.4.4 TOPICAL AND MUCOSAL DOSAGE FORMS

Topical and mucosal dosage forms of the invention include, but are not limited to, sprays, aerosols, solutions, emulsions, suspensions, or other forms known to one of skill in the art. *See, e.g., Remington's Pharmaceutical Sciences*, 16^{th} and 18^{th} eds., Mack Publishing, Easton PA (1980 & 1990); and *Introduction to Pharmaceutical Dosage Forms*, 4th ed., Lea & Febiger, Philadelphia (1985). Dosage forms suitable for treating mucosal tissues within the oral cavity can be formulated as mouthwashes or as oral gels.

Suitable excipients (*e.g.*, carriers and diluents) and other materials that can be used to provide topical and mucosal dosage forms encompassed by this invention are well known to those skilled in the pharmaceutical arts, and depend on the particular tissue to which a given pharmaceutical composition or dosage form will be applied. With that fact in mind, typical excipients include, but are not limited to, water, acetone, ethanol, ethylene glycol,

propylene glycol, butane-1,3-diol, isopropyl myristate, isopropyl palmitate, mineral oil, and mixtures thereof to form solutions, emulsions or gels, which are non-toxic and pharmaceutically acceptable. Moisturizers or humectants can also be added to pharmaceutical compositions and dosage forms if desired. Examples of such additional ingredients are well known in the art. *See, e.g., Remington's Pharmaceutical Sciences*, 16^{th} and 18^{th} eds., Mack Publishing, Easton PA (1980 & 1990).

The pH of a pharmaceutical composition or dosage form may also be adjusted to improve delivery of one or more active ingredients. Similarly, the polarity of a solvent carrier, its ionic strength, or tonicity can be adjusted to improve delivery. Compounds such as stearates can also be added to pharmaceutical compositions or dosage forms to advantageously alter the hydrophilicity or lipophilicity of one or more active ingredients so as to improve delivery. In this regard, stearates can serve as a lipid vehicle for the formulation, as an emulsifying agent or surfactant, and as a delivery-enhancing or penetration-enhancing agent. Different salts, hydrates or solvates of the active ingredients can be used to further adjust the properties of the resulting composition.

4.4.5 KITS

Typically, active ingredients of the invention are preferably not administered to a patient at the same time or by the same route of administration. This invention therefore encompasses kits which, when used by the medical practitioner, can simplify the administration of appropriate amounts of active ingredients to a patient.

A typical kit of the invention comprises a dosage form of a selective cytokine inhibitory drug, or a pharmaceutically acceptable salt, solvate, hydrate, stereoisomer, prodrug, or clathrate thereof. Kits encompassed by this invention can further comprise additional active ingredients such as, but not limited to, interferon-α, hydroxyurea, anagrelide, arsenic troxide, ST1571, imatinib mesylate, DX-8951f, R115777, vincristine, daunorubicin, prednisone, or a pharmacologically active mutant or derivative thereof, or a combination thereof. Examples of the additional active ingredients include, but are not limited to, those disclosed herein (*see, e.g.*, section 4.2).

Kits of the invention can further comprise devices that are used to administer the active ingredients. Examples of such devices include, but are not limited to, syringes, drip bags, patches, and inhalers.

Kits of the invention can further comprise cells or blood for transplantation as well as pharmaceutically acceptable vehicles that can be used to administer one or more active ingredients. For example, if an active ingredient is provided in a solid form that must be

reconstituted for parenteral administration, the kit can comprise a sealed container of a suitable vehicle in which the active ingredient can be dissolved to form a particulate-free sterile solution that is suitable for parenteral administration. Examples of pharmaceutically acceptable vehicles include, but are not limited to: Water for Injection USP; aqueous vehicles such as, but not limited to, Sodium Chloride Injection, Ringer's Injection, Dextrose Injection, Dextrose and Sodium Chloride Injection, and Lactated Ringer's Injection; water-miscible vehicles such as, but not limited to, ethyl alcohol, polyethylene glycol, and polypropylene glycol; and non-aqueous vehicles such as, but not limited to, corn oil, cottonseed oil, peanut oil, sesame oil, ethyl oleate, isopropyl myristate, and benzyl benzoate.

5. EXAMPLES

The following studies are intended to further illustrate the invention without limiting its scope.

5.1 PHARMACOLOGY AND TOXICOLOGY STUDIES

A series of non-clinical pharmacology and toxicology studies are performed to support the clinical evaluation of selective cytokine inhibitory drugs in human subjects. These studies are performed in accordance with internationally recognized guidelines for study design and in compliance with the requirements of Good Laboratory Practice (GLP), unless otherwise noted.

The pharmacological properties of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide, including activity comparisons with thalidomide, are characterized in *in vitro* studies. Studies examine the effects of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide on the production of various cytokines. In addition, a safety pharmacology study of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro- isoindol-2-yl)-propionamide is conducted in dogs and the effects of the compound on ECG parameters are examined further as part of three repeat-dose toxicity studies in primates.

5.2 MODULATION OF CYTOKINE PRODUCTION

Inhibition of TNF-α production following LPS-stimulation of human PBMC and human whole blood by 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide is investigated *in vitro* (Muller *et al., Bioorg. Med. Chem. Lett.* 9:1625-1630, 1999). The IC_{50}'s of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3 -dihydro-isoindol-2-yl)-

propionamide for inhibiting production of TNF-α following LPS-stimulation of PBMC and human whole blood is measured.

5.3 TOXICOLOGY STUDIES

The effects of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide on cardiovascular and respiratory function are investigated in anesthetized dogs. Two groups of Beagle dogs (2/sex/group) are used. One group receives three doses of vehicle only and the other receives three ascending doses of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide (400, 800, and 1,200 mg/kg/day). In all cases, doses of 3-(3,4-dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide or vehicle are successively administered *via* infusion through the jugular vein separated by intervals of at least 30 minutes.

The cardiovascular and respiratory changes induced by 3-(3,4- dimethoxy-phenyl)-3-(1-oxo-1,3-dihydro-isoindol-2-yl)-propionamide are minimal at all doses when compared to the vehicle control group.

All patents cited herein are incorporated by reference in their entireties. Embodiments of the invention described herein are only a sampling of the scope of the invention. The full scope of the invention is better understood with reference to the attached claims.